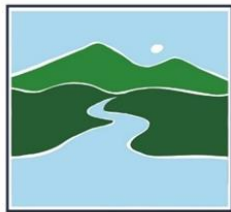


Watershed Management Division

Field Methods Manual



VERMONT DEPARTMENT OF
ENVIRONMENTAL CONSERVATION
WATERSHED
MANAGEMENT DIVISION

Vermont Agency of Natural Resources
Department of Environmental Conservation
Watershed Management Division
1 National Life Drive, Davis 3, Montpelier, VT
Vermont Agriculture and Environmental Laboratory,
163 Admin Drive, Randolph Center, VT
<https://dec.vermont.gov/watershed>
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1 Introduction

The Vermont Department of Environmental Conservation (VTDEC) [Watershed Management Division](#) (WSMD) is responsible for protecting, maintaining, and restoring the quality of Vermont's surface water resources. Integral to the success of this mission is monitoring the physical, chemical, and biological attributes of Vermont's 800+ lakes and ponds, 23,000 miles of rivers and streams, and 300,000 acres of wetlands.

The WSMD Field Methods Manual describes the equipment and procedures used to collect data on the status and trends of Vermont's surface waters. It is a resource for current and future field staff of WSMD, and for others seeking to collect data consistent with WSMD protocols. Many of the methods described here have been used for decades to monitor Vermont's waters, while others have been adopted recently as the result of advances in science and technology. The goal of the manual is to provide a recipe for data collection that is consistent, comparable, and scientifically rigorous. This version replaces the [2012 VTDEC Water Quality Division Field Methods Manual](#).

Many procedures describe protocols for use of specific field equipment and instruments. VTDEC does not endorse any particular brand, company, or model. Inclusion of equipment specific to certain brands, companies or models is simply a reflection of field gear currently used by WSMD staff.

2 General Guidelines

2.1 Safety

Work in the field and in the laboratory present unique health and safety risks associated with driving, boating, the use of specialized equipment, exposure to the elements and the handling of potentially hazardous chemicals. To mitigate these risks, all those conducting field work and/or laboratory work for the VTDEC WSMD are expected to read, understand, and abide by the general safety guidelines listed below as well as any safety policies that relate to specific types of work. These policies apply to all full-time, part-time, and temporary employees as well as interns, AmeriCorps members and volunteers engaged in field or laboratory operations.

1. All those driving to and from field sites must have a valid driver's license and have completed the Vermont State Defensive Driving course.
2. All those operating a motorboat must complete the [Vermont Boating Safety Course](#).
3. Fieldwork should be conducted in groups of at least two. Solo fieldwork should be avoided except in certain circumstances where safety risk is deemed negligible.
4. Field work in and around water creates risk of drowning and hypothermia. Proper protective equipment such as waders, personal floatation devices (PFDs), and survival suits should be utilized to reduce these risks.
5. Ticks are found throughout Vermont and are likely to be encountered. Field personnel are advised to follow [Vermont Department of Health Guidelines](#) on preventing tick bites and tickborne diseases.
6. All those regularly involved in electrofishing operations must follow the guidelines described in the VTDEC Safety Policy for Electrofishing Operations (VTDEC, 2018).
7. All those involved in laboratory work in the WSMD Laboratory at Vermont Agriculture and Environmental Laboratory (VAEL) must read and understand the WSMD Laboratory Chemical Hygiene Plan (VTDEC, 2021).

2.2 Preventing Biological Contamination

When conducting fieldwork, it is imperative to be aware of and take precautions to minimize the chance of contributing to the spread of non-native species and diseases that impact aquatic organisms. Aquatic non-native species such as Zebra mussels and Eurasian water milfoil are present in Vermont and pose a major threat to the integrity of aquatic ecosystems. Fish diseases such as Whirling disease are less extensive but when present in a waterbody (e.g., the Batten Kill) can be of great concern. The distribution of amphibian diseases such as chytrid fungus and ranavirus are less well-known but have been associated with large-scale die-offs. Precautions are necessary because there may be diseases yet to be discovered or established in Vermont. To minimize the risk for biological contamination between waterbodies, the following guidelines are to be followed:

1. Arrange field work so that un-infested sites are visited first and sites with known non-native species or aquatic diseases are scheduled at the end of the day.
2. Inspect all equipment prior to and after use. Remove mud, snails, algae, aquatic plants, and other debris from equipment if present. Equipment includes, but is not limited to nets, traps, boots, waders, float tubes, vehicle tires, boats, trailers, anchors, anchor chains and all sampling equipment. Allow gear to dry completely for 48 hours before being used again.
3. When visiting multiple sites in one day and allowing equipment to dry completely is not possible, a disinfecting solution should be used to ensure that biological organisms nor pathogens are transported between waterbodies. A disinfecting solution, such as Quaternary Ammonium (QUAT) or a 10% bleach solution should be thoroughly applied to sampling gear in between sampling sites, at a distance of at least 10 meters from the edge of the waterbody.
4. When working at sites with known or suspected nuisance species and/or disease problems, or when sampling populations of rare or isolated species, wear disposable gloves and change them between handling each animal (used disposable gloves should be retained for safe disposal in sealed bags.) Dedicate sets of nets, boots, traps, and other equipment to each site being visited. Clean and store them separately at the end of each field day.
5. Drain water from motorboats, leave drain plug out, and let sit for thirty days.
6. If a motorboat that was retrieved from an infested waterbody will be used less than thirty days in an uninfested waterbody, decontaminate the watercraft, engine, and trailer with a decontamination unit to flush the engine, and power spray the watercraft and trailer prior to use to kill zebra mussel veligers and other non-native larvae within the boat engine. A decontamination unit is available for WSMD use at the ANR Annex.

3 Physical Parameter Methods

3.1 Temperature

3.1.1 Field Thermometer

Calibration:

Field thermometers (non-mercury) must first be compared against the traceable National Institute of Standards and Technology (NIST) thermometer owned and maintained by the Agency of Agriculture at VAEL. The correction factor should be recorded on the upper end of the field thermometer. Reach out to the VAEL Director to coordinate thermometer calibration.

Procedure:

1. To determine the temperature, immerse traceable thermometer and field thermometer in the water sample for at least one minute.
2. Keep both thermometers immersed in the water while reading the temperature on the traceable thermometer.
3. Record temperature from the traceable thermometer to the nearest degree, then subtract the difference between it and the field thermometer.
4. Add the positive or negative correction factor to the field thermometer to insure accurate readings.
5. Field thermometers should be stored in a protective case when not in use.

3.1.2 Thermistor

Calibration:

Thermistors must be calibrated electronically every year by certified technicians. A two-point calibration bracketing the expected temperature range should be conducted. The probe line should be measured every year and re-marked if necessary.

Procedure:

1. Unwrap cable from the spool and lower the weighted probe into the water, allowing the cable to fully extend.
2. Turn the meter on and allow the reading to stabilize.
3. The probe line is marked in meters. Lower the probe to the desired depth, beginning at the lake surface if a profile is to be taken. Record the temperature in degrees Celsius, to the nearest tenth degree. Be sure the meter has stabilized before taking each reading.
4. When finished, turn the meter off, and rewind the cable.

Temperature is also measured in the field using multi-probes. See *Section 4.3 Measurements Using a Multiple Parameter Probe* for more information.

3.1.3 Continuous Temperature Sensors

Low-cost “continuous” sensors (also referred to as “loggers”) that record temperature at short, regular time intervals (e.g., every 15, 30, or 60 minutes) are now widely available. These loggers typically have a battery life of several years and can be deployed year-round. These high frequency measurements capture temporal patterns and episodic events that may otherwise be missed with limited discrete measurements. When used to monitor water temperature, these sensors can pattern spatial and temporal trends in stream networks, including the bracketing of suspected source areas of temperature stress. Pairing continuous air temperature data with water temperature measurements has utility for better understanding localized temperature dynamics, and for quality assurance purposes.

3.1.3.1 Sensor Accuracy

Before deployment, verify that temperature loggers:

- a. Have sufficient battery life and are launching and downloading data properly. Most sensors commence logging, and download subsequent data, using a data shuttle device that is synced with a computer. Data logging is typically set to begin under ambient field conditions, however testing that the sensors are working properly in the office or lab environment will help ensure proper field functioning of the loggers.
- b. Meet the minimum accuracy quoted by the manufacturer. Perform either single or multi-point accuracy checks by comparing sensor readings to a traceable calibrated thermometer. A commonly used single-point method involves placing sensors in an ice bath for several hours and comparing data from the sensor with a traceable thermometer.

3.1.3.2 Lotic Deployments

The information below is a brief synopsis of lotic deployment procedures and quality assurance protocols. A complete explanation of these procedures can be found in the USEPA’s guidance document *Best Practices for Continuous Monitoring of Temperature and Flow in Wadeable Streams* (USEPA 2014).

Procedure:

1. Evaluate location of lotic deployment using the following criteria:
 - a. Location is representative of the area where biological or other data is also being collected (if applicable).
 - b. Location represents well-mixed water.
 - c. Location is deep enough to keep the logger submerged for the duration of the deployment. Where feasible, water temperature loggers should be placed approximately 6 inches above the stream bottom. Loggers too close to the stream bed are susceptible to burial by moving substrates.
 - d. Location is protected from physical impacts associated with high flow events. If conditions permit, locate sensors downstream of a permanent structure (e.g., large boulder).

2. Water temperature loggers should be deployed inside radiation shields, as direct sunlight to the loggers can bias temperature readings. Housings made from polyvinyl chloride (PVC) are simple and easy to construct and offer protection for the logger. Housings should be of adequate size to easily hold the loggers, and numerous holes should be drilled in the housing to allow for water to circulate through the housing. Zip-ties can be used to secure the logger to the housing through these drilled holes.
3. Temporary water temperature logger deployments can be done by anchoring the logger and housing to an object or structure that cannot be dislodged during high flow events. Using a stainless-steel cable to attach the unit to a heavy cement block works well in most wadeable streams, with the cement block placed on the downstream side of a permanent structure if possible. The cable can be threaded through the cement block and PVC housing, and the housing should sit above the block to allow for water circulation (*Figure 3.1.A*). The PVC housing should be constructed such that the portion



Figure 3.1.A Sensor deployment utilizing a cement block.

to which the logger is secured can be unscrewed or otherwise removed from a portion attached to the block by the cable. When instream conditions are unsuitable for the placement of a cement block, other methods as described by USEPA (2014) can be used. These include cabling the housing to a length of rebar that has been driven into the streambed, or to a stable instream or bank structure.

4. Water temperature logger deployments can provide the most consistent data when the logger and PVC housing are permanently affixed to a stable instream structure using underwater epoxy. A PVC housing should be used whereby a portion of the drilled housing holding the logger can be unscrewed or otherwise removed from a permanently affixed portion attached to the structure. Once a suitable attachment surface is found (generally on the downstream side of the structure, in an area of well mixed water, at least six inches from the streambed), the attachment surface should be cleaned of biofilm.
5. A sufficient amount of epoxy is mixed and used to affix the PVC housing the attachment surface. Because it can take multiple days for the epoxy to fully set, it is necessary to

lean a large rock against the housing to hold it in place (*Figure 3.1.B*). USEPA (2014) provides further detail for successfully using this deployment method. Permanent housings should ideally be attached during late summer low flows, and during a period of time when significant increased stream flow is not expected for several days. Housings can be removed using a screwdriver or chisel to separate the epoxy from the instream structure.



Figure 3.1.B Permanent water temperature logger deployment using in-stream substrate and epoxy.

3.1.3.3 Lentic Deployments

Follow the Regional Lake Monitoring protocols currently in draft form with Tetra Tech and EPA.

3.1.3.4 Data Retrieval and Sensor Maintenance

1. When launching the logger, use a “delayed start” feature (if available) so that the sensor does not start recording until at least 60 minutes after it is in position and has time to stabilize to ambient field temperature.
2. For permanent deployments, it is recommended that data from loggers is downloaded a minimum of twice per year. Frequent downloads help to minimize data gaps resulting from equipment loss and ensure that loggers are replaced before the battery has expired.
3. When downloading data, try to ensure that no logger measurements are recorded after the removal of the logger from its housing and before the time when the data is downloaded. If data is retrieved, downloaded and re-deployed, the logger should be set to delay the start of recording by at least 30-60 minutes to ensure that the sensor has stabilized to ambient field temperature. If temperature data has been recorded before the logger was at ambient field temperature at deployment, or after the logger was retrieved, it is essential that this data is deleted from the final data set.

4. Make sure that the logger is set to record at consistent regular intervals, e.g., on the hour (xx:00), half hour (xx:30), or quarter hour (xx:15). The frequency of measurement will vary depending on data needs and usage, but an interval of 30 minutes or less is recommended for ensuring precision of some temperature data metrics (e.g., daily maximum, daily minimum). If water and air temperature loggers are being deployed together, the sensors should be set to record at the exact same time.
5. At each field visit during deployment, a discrete/instantaneous temperature measurement should be taken with a traceable calibrated thermometer as close as possible to the sensor and as close as possible to the time when the sensor is recording a measurement. Comparing logger data to this discrete measurement will help ensure accuracy in the continuous sensor data over time. Further data QA/QC will need to be done in the office to ensure that data collected during the deployment was of high quality.
6. During mid-deployment field visits and data logger downloads, the logger and housing should be gently cleaned of any debris or biofouling (e.g., algae, macroinvertebrates) that may affect logger performance or data quality.
7. A log should be kept that records relevant information from each field visit involving logger download and/or retrieval. At a minimum, this log should include field visit date, time(s) of logger retrieval and/or re-deployment, data interval collected, logger serial number, discrete/instantaneous temperature readings, and any other important information related to that specific visit.

3.1.3.5 Air Temperature Sensor Deployment

1. Place air temperature loggers as close to the water temperature sensor as possible.
2. Place air temperature loggers at a height of approximately six feet off the ground and out of direct sunlight.
3. If deployed in a forested riparian area, mount the air temperature logger to the north-facing side of a tree. Because vegetation can create radiation microenvironments, try to minimize the amount of other vegetation immediately near the logger. If suitable trees for attachment are not available, attach the logger to the north side of an existing or constructed stable structure.
4. Deploy air temperature loggers inside radiation shields, as direct sunlight to the loggers can bias temperature readings. Low price radiation shields for this purpose are available from multiple retailers or can be constructed as demonstrated in USEPA (2014).

3.2 Water Transparency

Multi-parameter sondes are frequently used to record in-situ measurements of turbidity (NTU) in streams, lakes, and wetlands. Follow procedures described in *Section 4.3 Measurements Using a Multi-Parameter Probe*.

3.2.1 Lentic Waters: Secchi Disk Transparency

On lakes and ponds, the most common measurement for water transparency is the Secchi disk. It consists of a 20 cm metal disk painted with alternating black and white quarters. This disk is attached to a line marked in meter increments. The optimum conditions for determining Secchi disk transparency are at mid-day on calm water. Ideally, a view scope is used to minimize effects of glare and waves. If a scope is used, readings are also taken without the scope as well and each type of reading is recorded. For Secchi instructions specific to the Vermont Laying Monitoring Program, please refer to the 2013 [Vermont Lay Monitoring Program Manual](#).

Procedure:

1. Lower the Secchi disk over the shaded side of the boat and remove sunglasses. Sun glare off the surface of the water interferes with accurate measurement.
2. Lean over the side of the boat with your face as close to the water's surface as possible.
3. Lower the disc slowly until it just disappears from view and note the depth. Then raise it back up until it is barely visible and note the depth again. The Secchi disk transparency is the deepest point at which you can still see the Secchi disk.
4. The line attached to the Secchi disk is marked in meters. Estimate the Secchi disk depth to the nearest tenth meter by reading the marked line at the water's surface. Some Secchi disks have reel meter tapes for more precise measurements.
5. If the Secchi disk is visible resting on the bottom, the actual Secchi disk transparency cannot be measured. It is important to note that the transparency was at least the depth to the bottom of the lake. Record the Secchi transparency as ">" the total depth or by placing a "B" to indicate that the Secchi hit bottom.

3.2.2 Lotic Waters: Transparency Tube

Transparency tubes can be used to measure water clarity and suspended solids in flowing waters. The tube allows the user to collect shallow water and determine its transparency and relative turbidity. It consists of a clear plastic tube, marked off in centimeters with a small Secchi disk at the bottom. A Tygon tube closure valve releases water through this tube at the bottom.

Procedure:

1. Fill the transparency tube with river or stream water, making sure to stand downstream of the sampling point.
2. Peer into the top of the tube. If the black and white painted Secchi disk is visible, record the transparency as ">" the total depth in cm.
3. If the painted disk is not visible, slowly release water through the valve at the bottom of the tube, until the disk is visible. Record the transparency in cm.

3.3 Stream Flow

Stream flow, or discharge, is defined as the volume of flow past a particular stream cross-section for a specific period of time, normally expressed in cubic feet per second (cfs). Conversion to metric units of cubic meters per second (cms) may be made by dividing by 35.31 cfs/cms.

3.3.1 Principals of Flow Measurement

See [Measurement and Computation of Streamflow](#) (Rantz et. al, 1982), [Discharge Measurements at Gaging Stations](#) (Turnipseed and Sauer, 2010a) and [Best Practices for Continuous Monitoring of Temperature and Flow in Wadeable Streams](#) (USEPA 2014) for more detailed technical guidance.

Stream flow measurement is most often made by velocity-area method, summing the products of the partial areas of the stream cross-section and their respective average velocities. The preferred calculation method uses the USGS mid-section formula (Turnipseed and Sauer, 2010a).



1. Crews will select a cross section to optimize the following characteristics:
 - a. A relatively straight stream channel with defined edges and a fairly uniform shape.
 - b. Limited vegetative growth, large cobbles, and boulders.
 - c. No eddies, slack water, or turbulence.
 - d. Average depths greater than 0.5 feet and velocities greater than 0.5 feet per second. Similar flow to that at any associated water level sensor (e.g., within the same reach as the sensor, with no tributaries or drainpipes located between the cross section and the gaging station).
 - e. If possible, large rocks and debris increasing flow complexity should be removed from the area of the section prior to the start of the measurement. After the section is modified, measurements should not be started until flow conditions have stabilized.
2. Hydraulically, at each subsection the point velocity at 60% of the depth (0.6d) below the water surface is approximately equivalent to the average velocity in the vertical velocity profile for depths equal to or below 1.5 feet. For depths greater than 1.5 feet, the average of two measurements at 0.2 and 0.8 of the total depth are more representative of total average velocity of the vertical. Place the measuring rod and meter at these location in the water column accordingly.
3. These measurements represent the average velocity for a subsection starting midway between the last location of the meter and the present location and ending midway to the next location. Remember the subsection limits when deciding where to position the rod for representativeness when irregularities in the streambed (e.g., boulders) are encountered.

4. Measure as many subsections as is practical to get reasonably accurate results. The number and lateral spacing of measurement verticals are selected based on the site-specific flow conditions with the goal of including no more than 10 percent of the total flow in any individual subsection. Therefore, readings should be closer together where there are greater depths and/or velocities. In general, 20-30 measurement locations should be enough to achieve this target.
5. It is good practice to periodically make two discharge measurements using two different cross sections of the stream as a check on accuracy.
6. Current meters and acoustic doppler velocimeters can be used effectively in most stream situations and in canals and flumes; however, if the velocities and/or depths are too low or the streambed too irregular, alternate techniques should be used in order to achieve a reasonable level of accuracy. Weirs, flumes, or volumetric techniques (i.e., a bucket or graduated cylinder with a stopwatch) should be used.
7. Occasionally, shallow, or slow velocity conditions may be dealt with by physically constricting the flow in order to increase depths and/or velocities.
8. Non-wadeable conditions on large rivers or during high flows may dictate measuring velocities throughout a cross-section by alternative methods, such with as meters and sounding weights suspended from a bridge, with an acoustic doppler current profiler or even surface floats. In the case of the latter, accuracy will be greatly reduced but may be the only alternative given conditions and available resources. See references for additional information on employing these methods.
9. Discharge measurements in streams under ice cover warrant additional considerations and techniques. See references for details.

The SonTek Acoustic Doppler Current Meter - Flowtracker unit will be the primary equipment for collecting discharge measurements and used in the field as described by Section 5 of the FlowTracker Handheld ADV® Technical Manual (SonTek 2009). Data processing and download will be performed per Section 6 of the Operation Manual.

3.3.2 Rating Curve Development

Discharge measurements at permanent streamflow gaging stations are collected over a range of flows to develop stage-discharge rating curves, which will allow water level measurements to be converted to discharge (see section 3.4 *Measurement of Water Level*). To establish a rating curve at a new site, a minimum of 5 to 10 discharge measurements will be made at a variety of stages, covering as wide a range of flows as possible. The rating curve will be produced by plotting instantaneous flow measurements over a range of stage heights. Other scientifically defensible methods, such as the slope-area or slope-conveyance (Rantz et al., 1982) for defining the high flow portion of a rating curve may be used where direct measurement is not possible. After establishing a rating curve, discharge will be measured monthly and also after large storms or any other potentially channel-disturbing activities, to verify or (if needed) update the curve. If new measurements are more than 15% off the rating curve, follow-up measurements will be made to identify whether a shift has occurred and, if necessary, to perform rating curve shifts or establish a new rating curve.

3.4 Measurement of Water Level

Water level, or stage, is measured continuously at streamflow gaging stations relative to reference benchmarks to define the site-specific relationships between stage and discharge and develop time-series of continuous streamflow (hydrographs). Methods follow those of EPA, 2014 and portions of Sauer and Turnipseed (2010b). See these for additional detail. Similar methods are periodically employed for automated water level monitoring in lakes, ponds, and reservoirs. Unvented submersible pressure transducers are the most often used sensor for measurement and logging of water levels. The following is a summary of the Watershed Management Division's methods:

1. Install instream pressure transducers in pools or deeper runs that optimize the following characteristics:
 - a. The water level data should be representative of the characteristics of the reach from which the biological data were collected. Note that for general monitoring purposes, the sensor need not be placed in the exact location where biological data are being collected.
 - b. Ensure that the site is not in the immediate vicinity of tributaries entering the river, and that no water is entering or exiting between the pressure transducer and the biological sampling site (e.g., through tributaries, pumping, or diversions). The goal is to minimize potential impacts from backwater during high flows (tributary downstream) or unevenly distributed streamflow across the channel (tributary upstream).
 - c. The gaging equipment should be installed in a pool where turbulence is minimal to increase accuracy of gage and transducer readings. The pool should have a downstream control feature that allows for stable stage measurements and ensures that the equipment will be submerged during low flows. Natural controls might include a downstream riffle, bedrock outcrop, or other stream feature that controls water levels. Unnatural controls might include a bridge or culvert that is narrower than the stream channel and constricts flow. Note that the feature controlling the stage-discharge relationship can change at different flow levels; such changes will be reflected in the rating curve.
 - d. To the extent practicable, a site should avoid beaver activity, extensive aquatic vegetation, or unstable streambeds and banks. These factors can change or result in shifting stage-discharge relationships.

More detailed information on site selection and controls can be found in Rantz et al. (1982) and Sauer and Turnipseed (2010b).

2. Two installation techniques for pressure transducers are generally relied upon: the fixed object method, in which the transducer and housing are attached vertically to a staff gage board or to an object like a bridge, boulder, or weir; and the streambed/bank method, in which the transducer and housing are affixed horizontally to the streambed or at an angle to the stream bank with longer housing pipe and held in place by hardware with rebar driven several feet into the bed or bank. Site-specific conditions dictate which technique is most appropriate. Wherever possible, stable staff gages are also installed in-stream for convenient measurement of reference water levels.

3. **Elevation Surveys:** Elevation surveys will be performed at sites with pressure transducers, using survey equipment that is maintained and operated in accordance with manufacturer specifications. Elevation surveys of the staff gage (if applicable), the current water level, and a fixed point on the in-stream gage structure will be surveyed relative to local benchmarks, with time and date noted. These elevation surveys allow for “reference water levels” against which logged depths are adjusted and stage-discharge ratings developed and maintained. During subsequent site visits reference water levels are easily noted from staff gage observation (where they exist). A benchmark and one or two other permanent secondary reference markers will be established at each site. The benchmark serves as the predominant reference point while the permanent reference marker(s) provide a backup in case the benchmark is destroyed and allows for a check of benchmark movement. Ideally, at least one reference marker should be located outside of the floodplain. Examples of good reference markers include: a bolt installed in a bridge, tree, bedrock, large boulder or utility pole. When in-stream benchmarks allow, reference water level is sometimes measured off benchmarks using carpenter T-square and torpedo bubble level. Where identifiable, the elevation of the lowest in-stream point on the primary downstream section control is surveyed. This provides the “gage height of zero flow” for later use in streamflow rating curve development and maintenance. Elevation surveys will be performed when new pressure transducers and staff gages are installed. After the initial survey, surveys will be performed at least once a year to identify if and when movement occurs. This is particularly important after high flow events and periods of extended ice cover. Once the survey is completed successfully, the elevation of all survey points will be compared to previously surveyed elevations to determine if movement occurred (sites are considered stable if elevations differ by less than 0.02 ft. If movement has occurred, adjustments will be made to the data or gage, as appropriate.
4. After the initial deployment, sites are visited within the first two weeks and then approximately monthly to check the condition of the sensors, gather data for mid-deployment accuracy checks and offload data. Where cellular telemetry is possible, data will be accessed more frequently to verify proper function and as needed in response to any QC alarm notifications. More frequent site visits help ensure the longevity of the sensors and data quality, and capture flow measurements over a range of water levels. Records of these checks will be kept on field notes. Typical maintenance checks will include:
 - a. Looking for signs of movement, physical damage, vandalism, or other disturbance, particularly after high flows, floods, and periods of ice cover. If movement occurs and the elevation between the staff gage and the transducer changes (e.g., due to sediment accumulation or scour), the equipment is secured (if necessary) and resurveyed. Any differences in gage and transducer elevation are noted on the field form. If a sensor is constantly shifting, an alternative location is considered (and if the sensor is moved, the new location is documented on the field form).
 - b. Ensuring that the (in-water) sensors are submerged. If the sensor is dewatered, this is noted on the field form. If the sensor is consistently dewatered, an alternative location is considered (and if the sensor is moved, the new location is documented on the field form).
 - c. Ensuring that the sensors are not buried in sediment. If the sensor is buried, this is noted on the field form and the sediment is removed. If the sensor is

- consistently buried in sediment, an alternative location is considered (and if the sensor is moved, the new location is documented on the field form).
- d. Removing anything that could bias the measurements. This includes clearing leaf litter and debris, which may pile up against the staff gage, transducer, and downstream control. This material is cleared at the beginning of a site visit, as it could impact gage height. The stage before and after debris clearing is noted on the field form. Stage data may be corrected if changes are detected.
 - e. Cleaning sediment or algae from the sensors. These can cause fouling and inaccurate readings. Sensor manuals are consulted for specific instructions on cleaning and maintenance.
 - f. Checking the condition of desiccant packets (vented pressure transducers only). The vented cable needs to remain dry in order for the sensor to function properly. Different transducers use different types of packets, and the lifespan of these packets varies depending on site-specific conditions (e.g., how much moisture is present in the air).
 - g. Ensuring that moisture is not filling the bottom of the canister that houses the open-air, non-vented pressure transducer. If moisture inundates the ports through which the barometric pressure is compensated, the measurements will be inaccurate.
 - h. Cleaning the staff gage with a scrub brush (especially during the summer months) so that the gage can be accurately read. Rust marks are painted over with enamel paint to improve durability.
 - i. Checking the battery life of the sensor (if appropriate).
 - j. Taking thorough field notes and photos to document any changes to the monitoring location (particularly those that could influence readings).
5. Sensor drift and logging error detection: Pressure transducer readings can drift over time, which can result in deviation of transducer data from that observed at the staff gage. Making periodic staff gage readings or benchmark checks to detect any shift or drift in the transducer data is important. The stage will be recorded every time the site is visited for a discharge measurement or data download. Gage readings will be done at least every month, to ensure a variety of stages are captured throughout the deployment period. Frequent gage readings facilitate error screening and early detection and correction of transducer problems that help minimize data loss. Pressure transducers might record erroneous readings during deployment for a variety of reasons, including:
- a. low flow conditions resulting in dewatering
 - b. high flow events that bury them in sediment
 - c. high flow events that move them
 - d. fouling from debris, aquatic vegetation, or algae
 - e. human-caused interference
 - f. becoming encased in ice
 - g. moisture entering the cable of a vented transducer, resulting in erratic readings or readings of zero water depth
 - h. the cable of a vented transducer becoming kinked or plugged, resulting in the data not being corrected for barometric pressure
6. Because these errors may occur, data will be screened. A series of error screening checks is described in Appendix K of EPA (2014).
7. Operation and data management: Water level loggers are operated, calibrated and data processed as described by manufacturer's recommendations, e.g., Onset (2017a).

3.5 Precipitation Measurements

The HOBO® Data Logging Rain Gauge, Model RG3 rain gauge is used occasionally to measure rainfall volumes during non-freezing conditions for special studies and watershed modeling.

Equipment:

- Tipping bucket rainfall collector
- HOBO Pendant® Event/Temperature Data Logger
- HOBO® Data Shuttle
- rigid post, tripod, or mountable horizontal surface
- bubble levels
- shims
- washers and wood screws for mounting and leveling bucket
- hose clamps
- HOBOWare® software program

Operation and data management:

The RG3 rain gauge will be operated and data processed as described by its user manual as well as the HOBO® Pendant® Event Data Logger Manual (Onset, 2017).

Installation Procedure:

1. The accuracy of all rain gauges is susceptible to exposure and wind effects, so care is taken to minimize impacts of wind and surrounding trees/buildings that may hinder the catch efficiency of the rain gage.
2. The height above ground and surrounding objects will be sufficient to eliminate the potential for splashing of nearby raindrops into the bucket. Typical guidance includes installation with rim of gage within 2-5 feet of the ground and in a location where the distance-to-height ratio of surrounding objects is ideally 2:1, or at a minimum 1:1.
3. Gauges are mounted to a sturdy object to avoid errant tips caused by vibration, and so that the opening is level in all directions as measured by a bubble gauge. Preference is given to securing on top of a horizontal surface, or gauges are affixed vertically to a post or pole with hose clamps. Where suitable mounting surfaces at sites do not exist, tripods anchored by driving rebar into the soil at an angle through the feet, sturdy fences, or posts driven securely into the ground are used for deployment. If mounting to a post, cutting the top of the post at an angle sloping away from the gauge reduces water splashing into the gauge.
4. Level positioning can be achieved more precisely with additional washers placed under one or more of the gauge feet if mounted on top of a horizontal surface, or with shims if attached vertically to a post.

Calibration Procedure:

1. A very small hole (a pinhole) is made in the bottom of a plastic or metal container of at least one liter capacity.
2. Place the container in the top funnel of the Rain Gauge with the pinhole positioned so that the water does not drip directly down the funnel orifice.
3. Pour exactly 473 ml of water into the container. Each tip of the bucket represents 0.01 inch of rainfall.

4. If it takes less than one hour for this water to run out, then the hole (from step 1) is too large. Repeat the test with a smaller hole.
5. Successful field calibration of this sort should result in one hundred tips plus or minus two.
6. Adjusting screws are located on the outside bottom of the Rain Gauge housing. These two socket head set screws require a 5/64 inch Allen wrench. Turning the screws clockwise increases the number of tips per measured amount of water. Turning the screws counterclockwise decreases the number of tips per measured amount of water. A 1/4 turn on both screws either clockwise or counterclockwise increases or decreases the number of tips by approximately one tip. Adjust both screws equally; if you turn one a half turn, then turn the other a half turn.
7. Repeat steps 3-6 as necessary until the Rain Gauge has been successfully calibrated. Perform calibration annually.

3.6 Physical Habitat Characterization - Lotic

Physical habitat characteristics are observed and recorded to supplement biological sampling data collected using lotic semi-quantitative macroinvertebrate methods (i.e., Section 6.5.1) and to help understand impacts to aquatic biota in general. The Ambient Biomonitoring Program (ABN) uses these supplemental data to further inform biological assessments. Additional information pertaining to the sampling site and samples collected are also noted. In general, all physical parameters are measured using the metric system, except for streamflow. The following information is recorded at each lotic biological monitoring site on paper field forms (Appendix B) or on electronic field forms.

3.6.1 General Information

3.6.1.1. Site Event Information

- Site (river) name and river mile (measured in distance from the mouth of the river in tenths of a mile)
- Site Location ID, if applicable, to ensure field data is matched with the correct site
- Date of sampling
- Time of sampling
- Latitude and Longitude (for new sites)
- Crew member names
- Site comments (directions, access instructions, description of site)
- Weather comments
- Photos of site using cell phone or tablet camera

3.6.1.2 Water Chemistry Sample Information

- Water chemistry sampler name
- Chemistry sample VAEL (WinLIMS) Customer Sample ID, if applicable, to ensure accurate tracking of water chemistry samples collected in the field.
- List of chemical parameters collected for analysis at VAEL
- Meter used (for in situ water chemistry measurements)
- Air temperature estimate (deg F)
- In situ water chemistry measurements (water temperature in Celsius, pH, Specific Conductivity (uS/cm), Dissolved Oxygen (% and mg/L), Turbidity (NTU))
- Estimated flow/precipitation within the past 48 hours. Note type of precipitation and, if known, how much. Local weather station and/or rain/flow gage data should be included if accessible.
- Estimated flow/precipitation within the past 2 weeks (note any prevailing weather patterns such as particularly wet or dry, ongoing drought, etc.)

Procedures for in situ water chemistry measurements are described in section 4 and collection of water samples are described in section 5.

3.6.1.3 Qualitative Flow Observations

In lotic waters, flow conditions are needed to accurately interpret water chemistry results. In the absence of a quantitative discharge measurement taken at the time of water sample collection, the water chemistry sampler should record a two-part qualitative streamflow observation: flow type/category and flow level.

- Flow Type (Base, Freshet or Hydro)
 - **Base Flow:** Stream flow is at a relatively constant level at the time of sampling, not rising nor dramatically falling in direct response to a rainfall event or snow melt runoff. Subsurface flows account for almost all water reaching streams. The hydrographs of nearby gaged streams have not begun to rise, have fallen to a similar level of that before the flow level rise began, or have leveled off to a steady but higher flow level. A base flow can exist under both low and sometimes moderate flows, but not under a “high” or “flood” streamflow level. The USGS maintains real-time streamflow data at <https://waterdata.usgs.gov/vt/nwis/rt>. This map and the hydrographs of current and recent conditions can be a helpful tool in identifying baseflow conditions.
 - **Freshet Flow:** During a freshet flow, a stream is actively rising or falling in response to a rain event or snow melt. The hydrograph of a nearby gaged stream shows an increase in flow, has not leveled off to the pre-event flow levels or stabilized to slightly higher than pre-event levels. Streams can be turbid under these conditions due to stormwater runoff and increased re-suspension of stream bed sediments.
 - **Hydro Flow:** During hydro flows, the stream’s flow level is rapidly rising or falling solely due to the release of water from an upstream dam. A rise in streamflow with no recent precipitation or snowmelt events and when similar rises are not observed for local stream gages are good indicators of artificial releases from dams. The Vermont Natural Resources Atlas, available at <http://anrmaps.vermont.gov/websites/anra/>, contains a *watershed protection* layer depicting known dams throughout the state, including whether they are operated for generation of electricity. The occurrence of natural freshet flows in direct response to rainfall or snowmelt are possible below such facilities.
- Flow Level (Low, Moderate or High)
 - **Low:** Streamflow conditions are believed to be low relative to the entire range of flows experienced at the site. If continuous stream gage data are available, streamflow conditions are generally expected to be greater than or equal to these levels 75% of the time (>Q75). Such low flows often occur during the late winter (January-February) and late summer (July-September). The streambed is often partially dry with channel bars exposed and it is possible to walk along the edge of a dry streambed.
 - **Moderate:** The stream is believed to be at a mid-level or average streamflow condition. These conditions exist approximately 25-75% of the time (Q25-75). This level can occur at any time of the year and are the most typical flows experienced in the stream. Approximately 90-100% of the stream bed is under water, and the stream bed will be almost fully wetted, but not adjacent to the sharp incline of the stream bank.
 - **High:** The stream is well-above an average level of flow. Streamflow conditions are generally expected to be greater than or equal to these levels only 25% of

the time (<Q25). These flows generally occur for extended durations in the spring and fall but occur for shorter periods of time in direct response to large rain events at any time of year. The stream may be full from bank to bank (“bankfull flows”) but is neither over its banks nor spilling onto the floodplain along most of its course. This streamflow level is never considered a “base flow” (see above).

- **Flood** – The stream is experiencing “flood” conditions, as indicated by water levels exceeding bankfull elevation and accessing the floodplain (if a well-defined floodplain exists at the site). Should there be no obvious floodplain feature adjacent to the channel, submergence of terrestrial and woody vegetation or active transport of large woody debris are other indicators of flood conditions. Flows of this magnitude are generally expected to occur less than 5% of the time.

3.6.2 Habitat

3.6.2.1 Physical Characteristics

- Habitat Type: (Riffle, Meandering Low Gradient, or Run) from which aquatic biota were sampled.
- Bank Stability: a qualitative rating (Poor, Fair, Good, Very Good or Excellent) based on streambank condition within sampling reach. Visually assess the stream banks and make note of undercut banks, erosion, rip rap, vegetation levels, bank steepness, and other factors.
- Velocity Class (Slow < 0.4 ft/s, Medium 0.4-2 ft/s, Fast >2 ft/s) based on visual estimate.
- Bankfull Width (m): measure or pace off the distance from bank to bank at a transect representative of the average bankfull width in the reach.
- Wetted Width (m): measure or pace off the wetted width of the stream at time of sampling, at a transect representative of the average wetted width in the sampling reach.
- General habitat comments and observations: Note any additional observations pertaining to physical habitat, stream condition, local land use or watershed characteristics at the time of sampling. Examples include odors, sheens, discolorations, activity on or near the stream bank, discharges or recent activity within the watershed, potential sources of pollution, etc.

3.6.2.2 Riparian Vegetation Structure

- Left Riparian Width (m): on left-hand side when facing upstream, visually estimate the average width of forested or otherwise naturally vegetated riparian zone perpendicular to the sampling reach, up to a maximum of 25 meters. Actively managed vegetation (i.e. crops, lawn, pasture) should not be included.
- Right Riparian Width (m): on the right-hand side when facing upstream visually estimate the average width of forested or otherwise naturally vegetated riparian zone perpendicular to the sampling reach, up to a maximum of 25 meters. Actively managed vegetation (i.e. crops, lawn pasture) should not be included.
- Softwood Overstory: visual estimate of the percentage of the 25-meter riparian zone overstory (> 5m high) composed of softwoods.
- Hardwood Overstory: visual estimate of the percentage of the 25-meter riparian zone overstory (> 5m high) composed of hardwoods.
 - Notes on characterizing the overstory:

- The percentages of softwood and hardwood do not need to add up to 100%, unless there is 100% overstory in the 25-meter riparian zone comprised of both softwoods and hardwoods.
- Overstory composition should be expressed as a percentage of a 100% forested riparian zone, regardless of whether the riparian zone is 100% forested.
- Example: if there is a fully forested 25-meter riparian zone comprised of hardwood trees on the right and there is a pasture with no overstory on the left, hardwood overstory would be 50%.
- Shrub Understory: visual estimate of the percentage of the 25-meter riparian zone understory (< 5m high) covered by woody shrub vegetation.
- Herbaceous Understory: visual estimate of the percentage of the 25-meter riparian zone understory (< 5m high) covered by herbaceous vegetation.
- Lawn/Pasture Understory: visual estimate of the percentage of the 25-meter riparian zone understory (< 5m high) covered by maintained lawns or pastures.
 - Note on characterizing understory – The percentages of shrub and herbaceous/lawn understory can equal more than 100%, with the understanding that shrub and herbaceous cover can overlap in highly productive riparian areas.
- Canopy: the percent of shading from overhanging riparian vegetation, estimated as an average at the mid-channel water surface throughout the sampling reach.
 - Visual Estimate Method - Stand in the center of the stream or river, extend both of your arms horizontally to each side creating a 180-degree angle. Observing the overhead canopy cover water surface level, start to lift your arms up from the horizontal position slowly towards your head. Stop when each arm is in alignment with the overhead canopy. Then estimate the angle of your left and right arm. Combine the percent canopy values from your left and right side to obtain the total percent canopy (Figure 3.6.A).

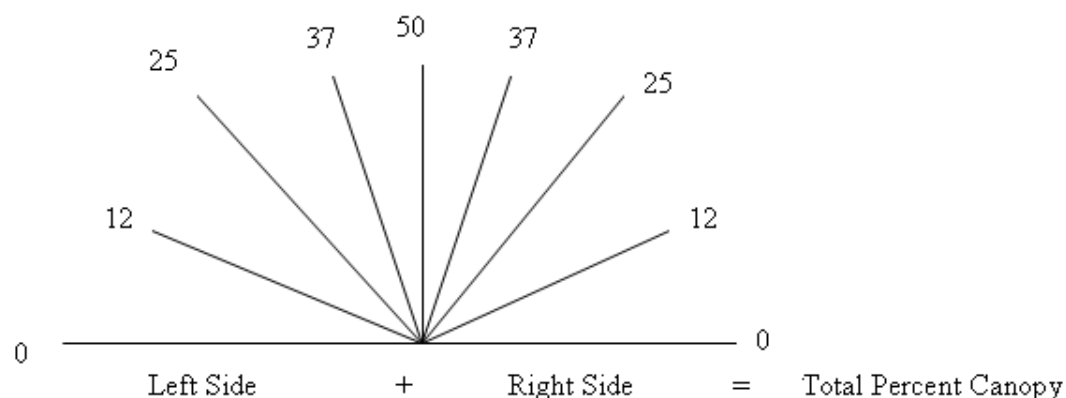


Figure 3.6.A Percent canopy determination

- Measurement Using Spherical Convex Densiometer
 - a. At mid-channel facing upstream, hold the densiometer 0.3 meters (~1 ft) above the water surface. Hold the densiometer so that it is level using the bubble indicator. Count the number of points covered by vegetation. Percent

canopy cover is estimated as number of points with vegetation divided by the total number of points, multiplied by 100. Record this value:





- i. $\% \text{ canopy} = (\# \text{ points with vegetation} / \text{total } \# \text{ points}) \times 100$
- b. Repeat the procedure above by facing downstream. Repeat measurement standing on the transect with the densiometer halfway between middle of wetted channel and the left bank. Repeat this procedure for the right bank.
- c. At this point you will have four measurements for the transect: two from the center and one halfway to each bank. Canopy cover is estimated as the mean of the four measurements.

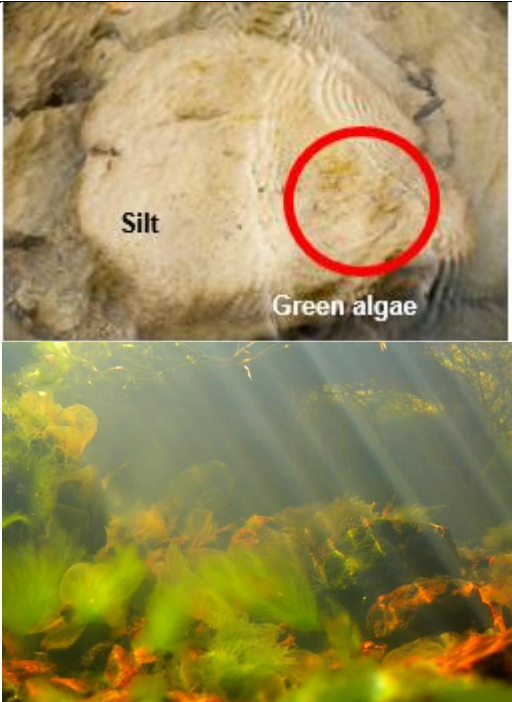


3.6.2.3 Substrate Characteristics

- **Embeddedness Estimate:** visual estimate of the extent to which coarse gravel and cobble sized substrate are buried in silt, sand, or fine gravel in a riffle area expressed as a percentage. As coarse substrates become embedded, the surface area available as habitat to macroinvertebrates and fish decreases, due in part to decreasing interstitial space. This estimate is best assessed after a pebble count or collecting a macroinvertebrate sample, when many coarse gravel or cobble pieces have been removed and examined. If a cobble is easily removed, embeddedness is minimal; if it is difficult to remove, it is high.
- **Silt Rating:** visual estimate on a scale of 1 (low) – 5 (high) silt prevalence in riffle sampling reach.
 - Physically disturb riffle by kicking up substrate, mimicking a heavy storm/high flow event.
 - Observe plume of debris dislodged in stream channel and assign a rating.
 - Signs of high silt include grey or light brown plume of fine particles that remain suspended in water column for > 3-5 seconds before dispersing.
 - When present, fine particulate organic matter (FPOM) is also dislodged when substrates are disturbed but should not count towards the silt rating. FPOM is typically darker and more brown or black in color, and may disperse more quickly than silt.
- **Iron Precipitate:** visual estimate of percentage of substrate surfaces with reddish orange iron precipitate.
- **Calcareous Deposits:** estimate of percentage of substrate surfaces covered in gritty, white-gray calcareous deposits, best assessed after pebble count.
- **Large Woody Debris:** number of large woody debris (> 0.3m in diameter) within the bankfull stream channel expressed as #/100m.

3.6.2.4 Periphyton Cover

Visually estimate percentage of surface of substrates within sampling reach covered by the following categories of periphyton: moss, filamentous green algae, blue green algae, biofilm, green algae, other periphyton and macrophytes.

Type	Description	Example
Moss	Short, stout patches or mats of small, thick stems. Rough in texture. Moss may be above or below the water surface.	
Filamentous green algae (macroalgae)	Stringy growth in patches or dense mats. Length can vary and can grow beyond the extent of substrate. Easy to break or pull apart. Usually green in color.	
Blue green algae (macroalgae)	Dark green/bluish splotches forming mat on substrate surface; sometimes appears brown as it dies off. Earthy smell.	
Biofilm (microalgae)	Biofilm layer covering substrate, typically lacking visibly discernable clusters or units and without green or brown color. Can be very thin (mildly slimy) or thick.	

Green algae (microalgae)	<p>Can be green or if starting to die back, brown. Lacking discernible stems or units, not as easily dislodged or as fine as silt. Lacks structural integrity of filamentous algae</p>	
Other periphyton	<p>Any other type of periphyton not listed here, such as didymo (pictured: thick, clumpy macro-algae that feels like cotton when rolled between fingers) or freshwater sponges.</p>	
Macrophytes	<p>Larger plants that are typically rooted in sediment vs. on substrate. Typically found in slower moving waters such as wetlands and slow winders. Riverweed is an exception, attached to substrate in fast flowing rivers.</p>	

3.6.3 Substrate Composition

3.6.3.1 Visual Estimate Method – Percent Composition

1. Locate a representative section of stream reach (riffle, meandering low gradient, or run). This will generally be the area where a macroinvertebrate sample has been collected.
2. Estimate the percentage of materials that make up the river bottom within the sampling reach. Estimate percentage of the following size categories:
 - a. Silt (smaller than a grain of sand; smooth texture when rubbed between fingers)
 - b. Sand <2 mm (<0.08" or smaller than a peppercorn); gritty
 - c. Gravel 2-16 mm (0.08-0.63" or peppercorn to marble size)
 - d. Coarse Gravel 16-64 mm (0.63-2.5" or marble to tennis ball size)
 - e. Cobble 64-256 mm (2.5-10.1" or tennis ball to basketball size)
 - f. Boulder >256 mm (>10.1" or larger than a basketball)
 - g. Ledge/exposed bedrock (larger than a car)
3. Visual substrate estimates are generally only taken in cases where modified pebble counts are not performed (e.g. low gradient streams).

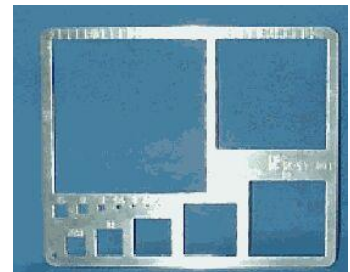
3.6.3.2 Gravelometer – Modified Pebble Count

This method is used to describe the substrate particle size classes within riffle habitats for macroinvertebrate community assessments. The method is based on the more rigorous technique developed by Wolmen (1954) to describe coarse riverbed materials. Modifications of this technique developed by the US Forest Service describe the channel bed materials within stream reaches (Beverger and King, 1995 and Harrelson et al, 1994).

The pebble count should occur after macroinvertebrate sample collection is complete to avoid disturbing substrate and impacting sample collection. Macroinvertebrate sample collection is described in section 6.5.

Equipment:

- Gravelometer: A metal plate with square openings ranging from 4 mm to 128 mm with notches on the top edge measure 180, 256 and 362 mm. Bottom notches provide additional measurements. The particle must be placed through the smallest opening so that the intermediate axis is perpendicular to the sides (not diagonally across) of the opening. The particle is classified by the smallest size category that the particle fits through.
- Field sheet (See Figure A.2) or electronic field form



Procedure:

1. Begin at the downstream end of the macroinvertebrate sampling reach.
2. Begin transect from edge of stream channel whether wetted or dry. At each observation point, avert the eyes away from the streambed to reduce visual bias on substrate

selected. Extend an index finger until it touches the first piece of substrate encountered in the streambed.

3. Determine the size category of the randomly selected substrate particle. Use the gravelometer or, if experienced, a visual estimate to determine size category according to the following biologically based size classes:
 - a. Silt (smaller than a grain of sand)
 - b. Sand <2 mm (<0.08" smaller than a peppercorn)
 - c. Gravel 2-16 mm (0.08-0.63" peppercorn up to marble size)
 - d. Coarse Gravel 16-64 mm (0.63-2.5" or marble to tennis ball size)
 - e. Cobble 64-256 mm (2.5-10.1" or tennis ball to basketball size)
 - f. Boulder >256 mm (>10.1" or larger than a basketball)
 - g. Ledge/exposed bedrock (larger than a car)
4. Pace off diagonal zigzag transects across the stream to cover the full length of the sampling reach, including dry gravel bars and substrate at the stream edge inside the bankfull width.
5. Select a pebble as described above at every two paces in larger streams (> 20 m bankfull width) or every pace in smaller streams (< 20 m bankfull width). Call out each particle size to the data recorder.
6. Examine 100 particles over the course of the reach.

3.6.3.3 Periphyton Cover Measurements

Periphyton cover should be measured at the same time as the Modified Pebble Count described above. Pebbles that are 16mm or larger (i.e. coarse gravel and larger substrates) can be assessed for periphyton cover. Pebbles smaller than 16mm (silt, sand, and gravel) cannot be observed for periphyton cover. Dry substrate should be measured for moss cover only but not for macro or micro algae. Notes should be taken that describe the nature of the periphyton communities within the sampling reach (e.g., moss mostly on the edges or mostly aquatic). Filamentous and blue-green algae are considered macro algae. Green algae and diatoms are considered micro algae. Periphyton should also be characterized on substrate that cannot be picked up from the streambed for observation. For larger substrate (e.g. boulders) that are only partially wetted, micro- and macro-algae cover should only be characterized from the submerged portion.

Procedure:

1. Follow steps 1-6 in Section 3.6.3.2: Gravelometer - Modified Pebble Count Procedure.
2. For each substrate selected with an intermediate diameter of 16mm or larger (i.e. coarse gravel and large substrates) record the amount of moss on a scale of 0-3, where:
 - a. 0 = no moss present
 - b. 1 = some (<5% coverage) moss present
 - c. 2 = 5-25% cover of substratum by moss
 - d. 3 = >25% cover of substratum by moss
3. For each substrate selected from within the wetted channel with an intermediate diameter of 16mm or larger (i.e. coarse gravel and larger substrates), record the amount of macro algae on a scale of 0-3, where:
 - a. 0 = no macro-algae present
 - b. 1 = some (<5% coverage) macro algae present
 - c. 2 = 5-25% cover of substratum by macro algae

- d. 3 = >25% cover of substratum by macro algae
- 4. For each substrate selected from within the wetted channel with an intermediate diameter of 16mm or larger (i.e. coarse gravel and larger substrates), estimate average thickness of micro algae on a scale of 0-5, where:
 - a. 0 = substrate is rough with no apparent microalgae present;
 - b. 1 = substrate is slimy, but biofilm is not visible (not thick enough to draw a line with the back of fingernail, endolithic algae can appear green but will not scratch easily from the substratum);
 - c. 2 = a thin layer of microalgae is visible (thick enough to draw a line in the biofilm with the back of your fingernail);
 - d. 3 = accumulation of microalgae to a thickness of 0.5-1 mm;
 - e. 4 = accumulation of microalgae from 1 mm to 5 mm thick;
 - f. 5 = accumulation of microalgae from 5 mm to 20 mm;
- 5. If didymo cover is present on substrates within sampling reach, record the amount in a separate count according to the categories described in #3 for macro algae.
- 6. If iron precipitates or calcareous deposits are present on substrates within the sampling reach, record the amount in a separate count according to the categories described in #4 for micro algae thickness.

3.7 Stream Geomorphic Assessment Lite

A Phase 2 SGA-lite has been developed to allow for a collection of the minimum amount of data needed to understand stream type and processes that guide appropriate restoration/protection projects and support other regulatory and monitoring efforts within the Division. Below is a list of the primary parameters necessary to gain an understanding of the general physical condition of a segment. Parameters and methods are based on the full [Phase 2 Stream Geomorphic Assessment \(SGA\) Protocols](#). Refer to the full Phase 2 SGA protocols for additional details and/or background information about the parameters measured in Phase 2 SGA-lite.

Primary Parameters: These parameters are considered essential to provide the minimum type and amount of data to base stream characterizations on and are needed to understand stream process and condition.

- Confinement
- Bankfull width
- Width/depth (W/D) ratio
- Entrenchment
- Channel type
- Incision
- Stream type
- Bed form
- Reach Geomorphic Assessment (RGA) – Geomorphic Condition
- Channel Evolution Model (CEM)
- Sensitivity

Where to Conduct a cross-section: Measurements of channel dimensions, such as bankfull width, maximum depth and flood prone width (Figure 3.7.1), are conducted at the channel "cross-over" locations (Figure 3.7.2). In a meandering stream, a cross-over is the area where the main current or flow in the channel crosses over from one side of the channel to the other. Riffles are usually located at cross-over locations. In steep gradient channels that run relatively straight, the main flow of the channel does not usually move from side to side across the channel, but rather cascades over cobble and boulder steps, or runs straight over a uniform channel bottom. In these stream types, channel dimensions should be measured at these cascades, steps, or uniform runs. In a very low gradient stream where distinct riffles may not form, you should still perform channel measurements at cross-over locations, which may resemble shallow runs. **Do not take channel measurements at pools or you will over-estimate the channel width and maximum depth. Be sure to mark on your sketch where you take cross section measurements.**

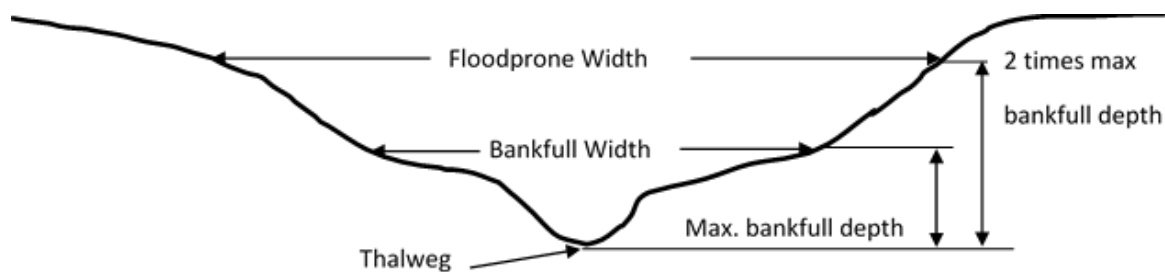


Figure 3.7.1 Channel dimensions - cross section view

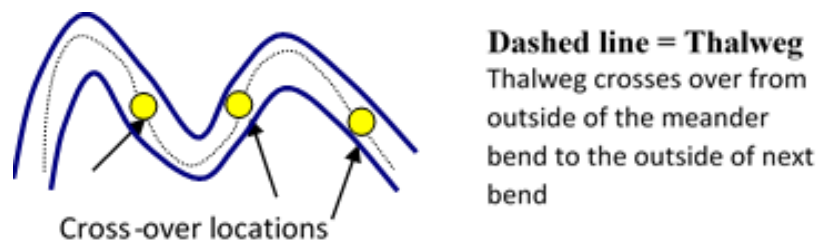


Figure 3.7.2 Cross-over locations - plan view

Recording Cross-section Data: A separate “[Cross-section Worksheet](#)” is used to record distance and depth measurements. If several cross-sections are completed do **not** record an average of channel dimensions at these cross-sections on the Field Notes form. Instead, record the set of values from the cross-section that is most typical of the segment (or reach). The goal is to capture the channel dimensions most prevalent throughout the segment (or reach). The Worksheet also provides an area for drawing and labeling a typical cross-section and for calculating stream bed particle size percentages at different bed features (from Pebble count). The information from this worksheet will then be transferred to an Excel workbook. For projects that have Phase 1 and/or Phase 2 Stream geomorphic assessment (SGA) projects, the Excel workbook will be uploaded to the [Stream Geomorphic Assessment Data Management System](#) for each reach. For projects outside of the Phase 1/Phase 2 SGA work, the Excel worksheet provides a way to store data for the users work and data evaluation. The Excel workbook provides a worksheet for entering several cross-sections for a segment. This is a good way of insuring that you have collected the needed information and confirm what you had calculated for channel information.

Setting up the cross-section:

It is important when doing a cross section to collect enough data to adequately characterize the relationship between the river, its floodplain and the valley. Make sure the cross section goes beyond the top of bank, and where feasible go out to the toe of the valley walls on either side of the channel. **Survey as much of the valley bottom as is reasonable keeping in mind the objective of depicting as complete a representation of the valley and channel morphology as possible given the time available**

Capturing features within the channel:

- a. **Bankfull width:** This measurement should be taken over a riffle or similar feature, such as a step, cascade, or run in steep channels. To measure bankfull width, stretch a

measuring tape taut across the channel, perpendicular to the flow direction, from the point of bankfull elevation on the left bank across the stream to the bankfull elevation on the right bank (Figure 3.7.3). Pin the tape at these two points at the bankfull elevation. View the stretched tape from downstream to be sure that it is level. Also check the levelness of your tape with a hand level if available, or use a measuring rod along the tape to make sure it is running equal distant from the water surface. Record the width to the nearest foot.

- b. **This feature may or may not be at the top of bank.** Be sure to leave a few blank spaces at the start of cross-section data sheet to collect data above and/or beyond the left bankfull pin for data on the left bank / floodplain.
- c. **Mean Depth:** While the tape is still stretched across the channel at bankfull elevation, use a depth rod to measure 10 depths at evenly spaced intervals across the channel (Figure 3.7.3). Average the ten measurements to determine a mean bankfull depth. The spacing interval used to measure depths across the channel is determined by dividing the bankfull width by 11. For instance, if the bankfull width is 50 feet, take a depth measurement approximately every 4.5 feet across the channel. At each interval record the distance across the channel (from the left bankfull pin) and the corresponding depth from the tape to the stream bed on the Cross-section Worksheet.
- d. **Max Depth:** While collecting the evenly spaced interval measures to obtain the mean depth, also collect the bankfull maximum depth. With the depth rod, find the deepest depth between the tape and the stream bed; this is called the thalweg (Figure 3.7.3). Record the distance from the left bankfull pin and depth from the tape to the streambed measurement to the nearest tenth of a foot.
- e. **Edge of Water:** In addition to getting the depths at the given intervals, capture the depth (from the tape to the streambed) at the left and right edge of water.
- f. **Additional features to make note of (Figure 3.7.5):**
 - i. Right and Left Top of Bank. This may or may not be at the same elevation as the bankfull feature. If the channel has incised the top of bank may be above your bank full feature.
 - ii. Significant changes in slope along the bank and/or channel bed.

Capturing features outside the channel:

- a. **Recently Abandoned Floodplain (RAF):** Stretch a tape taut and level across the channel from the top of the lowest of the two banks to a measuring rod positioned at the thalweg (Figure 3.7.4). Record the height of the recently abandoned floodplain to the nearest tenth of a foot, which is the distance between the measuring tape and the streambed at the thalweg. Record at least one point out beyond the top of bank point to help with determining if the feature continues at the same elevation for some distance or changes slope within a given distance.

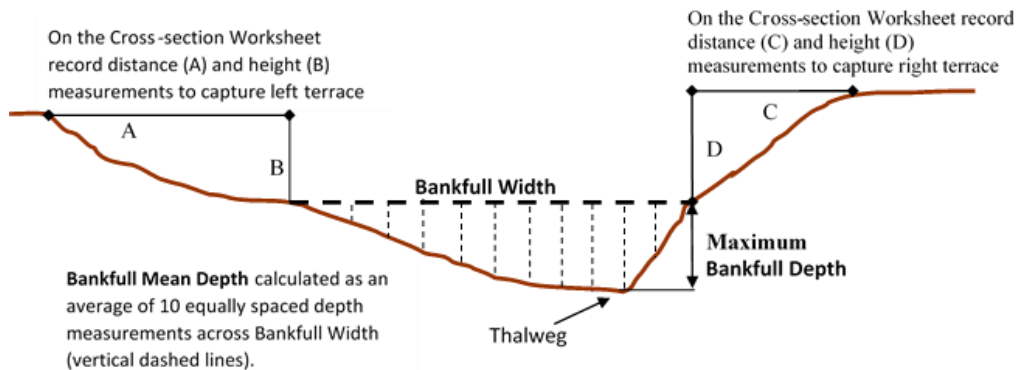


Figure 3.7.3 Diagram of bankfull width, maximum depth, mean depth and terrace measurements

- b. The RAF height is divided by bankfull maximum depth to determine the incision ratio for the channel. In some cases where the stream has not incised there will not be an abandoned floodplain and the bankfull elevation and the current floodplain elevation will be the same. If the bankfull maximum depth is identical to the height of the RAF then the same number will be recorded for both parameters on the Cross-section form. The incision ratio should be 1.
- c. **Floodprone Width:** The floodprone width is measured at an elevation that is 2 times the bankfull maximum depth (Figure 3.7.4.). For example, if the bankfull maximum depth is 3 ft., you would move the tape up to 6 ft. on the depth rod to reach the floodprone elevation. To measure your floodprone distance at the floodprone elevation, start at the bankfull pin, set the base of the depth rod at the bankfull elevation. Move the tape up the depth rod to height of the bankfull max depth. This is the same depth as 2 times the bankfull max depth if standing in the channel at the thalweg. Stretch the measuring tape out level across the adjacent floodplain until it intersects the next adjacent terrace or hillside at the floodprone elevation on either side of the channel. This total distance across the channel and floodplain area on both sides of the channel, measured at the floodprone elevation, is the floodprone width.

Record at least two elevations across the floodplain before encountering a terrace and/or valley wall feature. If the floodprone area is so wide that measuring it would take you far across the valley more than 500 feet, estimate the distance by eye or with a range finder, as this distance becomes impractical to measure with a measuring tape. Where there is large variability in the elevation across the floodplain, record elevations of features that are distinct across much of the floodplain; and not just a localized "bump" that may be caused by things like downed trees or random boulders. You are looking to capture features that would affect your flood depths and connection to floodplain areas.

- d. **Adjacent Terrace:** To capture adjacent terraces on either side of the channel (Figure 3.7.4.) record the elevation at the top of the break in slope and at least one point out beyond that point to help with determining if that feature remains at the same elevation for some distance or changes within a given distance. Be sure to leave space on your cross-section sheet to record these measurements.

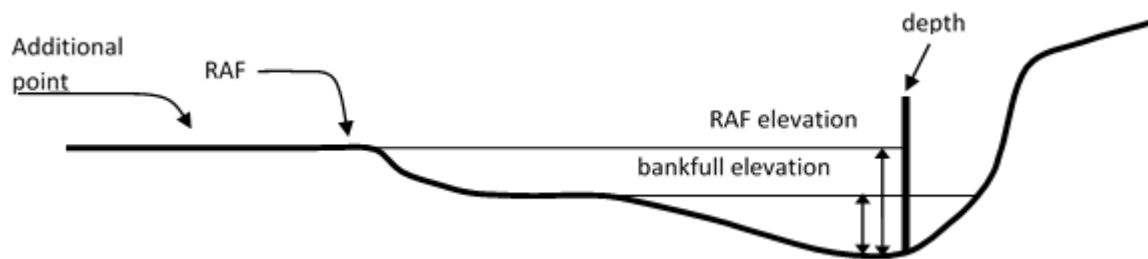


Figure 3.7.4 Measuring Recently Abandoned Floodplain (RAF)

Cross-section and Floodplain Calculations:

Width/ Depth Ratio: Divide the bankfull width by the **mean** bankfull depth . Example: If the stream has a bankfull width of 30 ft. and a mean depth of 2.0 ft., the width/depth ratio is $30 / 2 = 15$.

Entrenchment Ratio (ER): Can be a visual estimate based on the width of the valley at an elevation twice the bankfull depth compared to the bankfull width; or if measurements have been done, divide the width of the floodprone area by the bankfull width to determine the entrenchment ratio. Example: If the stream has a floodprone width of 100 and a width of 20 the entrenchment ratio is 5. See Table 3.7.A for a description and classification of the ER.

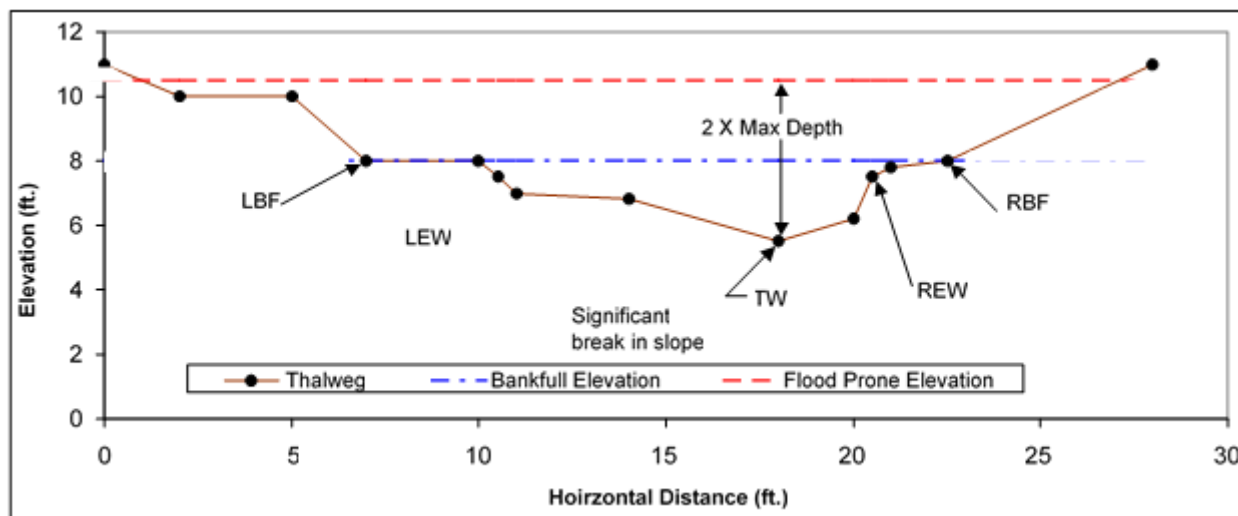


Figure 3.7.5– Example of Cross-section features to capture on field form.

Cross-Section Codes: LFPA = Left Floodplain, LTER = Left Terrace, LTOB = Left Top of Bank, LBF = Left Bankfull Stage, LEW = Left Edge of Water; TW = Thalweg, REW = Right Edge of Water; RTOB = Right Top of Bank, RBF = Right Bankfull Stage

Table 3.7.A. Table 3.7.A. Entrenchment Ratio Classification and Description

Entrenchment	Description	Ratio
Fully	Limited to No accessible floodplain beyond the channel banks	ER<1.4

Moderate	Minor to moderate floodplain area available beyond the channel banks. Accessible floodplain at higher flows	ER=1.4-2.2
Minor	Available floodplain area is more than twice the channel width beyond the channel banks. Generally, accessible floodplain at, or just above, the 1-2-year flow recurrence interval.	ER> 2.2

Incision Ratio (IR): This can be a visual estimate based on the comparison of the height of the top of bank compared to the bankfull height; or if measurements have been done, divide the recently abandoned floodplain (RAF) by the bankfull maximum depth. Values will always be greater than or equal to one. See Table 3.7.B for the classification and description of the incision ratio.

Table 3.7.B. Incision Ratio Classification and Description

Incision	Description	Ratio
Major	Sharp change in slope and/or multiple headcuts present, riffles/dunes/steps replaced by planebed features, extensive historic channel straightening, gravel mining, and/or recent channel avulsion, major existing flow alterations, greater flows and/or reduced sediment load	IR >2
Moderate	Sharp change in slope, headcuts present, riffles/steps/ dunes may appear incomplete/eroded, dominated by runs, evidence of historic straightening, dredging, gravel mining, and/or channel avulsions, major historic flow alteration, greater flows, and/or reduction in sediment load	IR ≥1.4 and <2
Minor	Minor localized slope increase or nickpoints, riffles/steps/dunes mostly complete but may appear shorter, evidence of minor historic dredging and/or channel avulsion	IR ≥1.2 and <1.4
Reference	Little evidence of localized slope increase or nickpoints, riffles/steps/dunes complete, no evidence of historic channel alteration (straightening, dredging) or avulsions. No known flow alterations	IR ≥1 and <1.2

Confinement: Can be a visual estimate (or coarse map measurement) based on ratio of bankfull width to valley width. Make note of whether there is a human caused change in confinement (for example, a road narrowing the valley). See Table 3.7.C. for the classification and description of confinement. Note this is different than Entrenchment. Confinement is comparing bankfull width to the width of the natural valley, or in the case of human encroachment, the human modified width. Entrenchment is the valley width available at 2 times bankfull max depth and may be narrower than the natural confinement ratio if the stream has become incised into the valley.

Table 3.7.C. Confinement Classification and Description

Confinement	Ratio = Valley Width / Channel Width
Narrowly confined	≥ 1 and < 2
Semi Confined	≥ 2 and < 4
Narrow	≥ 4 and < 6
Broad	≥ 6 and < 10
Very broad	≥ 10

Stream and Bedform Determination:

Stream Type: Using the measurements made in the steps above, and referring to Table 3.7.D. below, determine the existing stream type for the segment (or reach). Streams are placed into the different stream types based on their entrenchment, width-depth ratio, sinuosity, channel slope, substrate size, and bed features. If the stream type has been based on the use of the +/- factors allowed for entrenchment and width/depth; include why those factors were used in the comments.

Table 3.7.D. Stream Type parameters (1-3) in order of priority for typing (Rosgen 1994).

Stream Type	(1) Entrenchment Ratio (+/- 0.2 units)	(2) Width/depth (+/- 2 units)	(3) Sinuosity (+/- 0.2 units)	Slope % (See Note)
A – Single Thread	<1.4 - Entrenched	<12 – Low	<1.2 – Low	4-10
G – Single Thread	<1.4 - Entrenched	<12 – Low	>1.2 – Low to Mod.	2-4
F – Single Thread	<1.4 - Entrenched	>12 – Mod. to High	>1.2 – Low to Mod.	<2
B – Single Thread	1.4 -2.2 – Moderately Entrenched	>12 – Moderate	>1.2 – Low to Mod.	2-4
E – Single Thread	>2.2 – Slightly Entrenched	<12 – Very Low	>1.5 – Very High	<2
C – Single Thread	>2.2 – Slightly Entrenched	>12 – Mod. to High	>1.2 – Moderate	<2
D – Multiple Thread		>40 – Very high	<1.2 - Low	<4

Note: Channel slope is not a measured value in the Phase 2 protocol. Use an estimate of the channel slope as measured on a topographic map. Slope = change in upstream/downstream elevation of your reach (or segment) divided by the stream length.

Sinuosity:- Use Figure 3.7.6 to determine the sinuosity within your reach (segment), based on field observations and review of ortho-photos to describe the sinuosity of the bankfull channel. To calculate sinuosity = your reach (segment) length divided by valley length for your reach (segment).

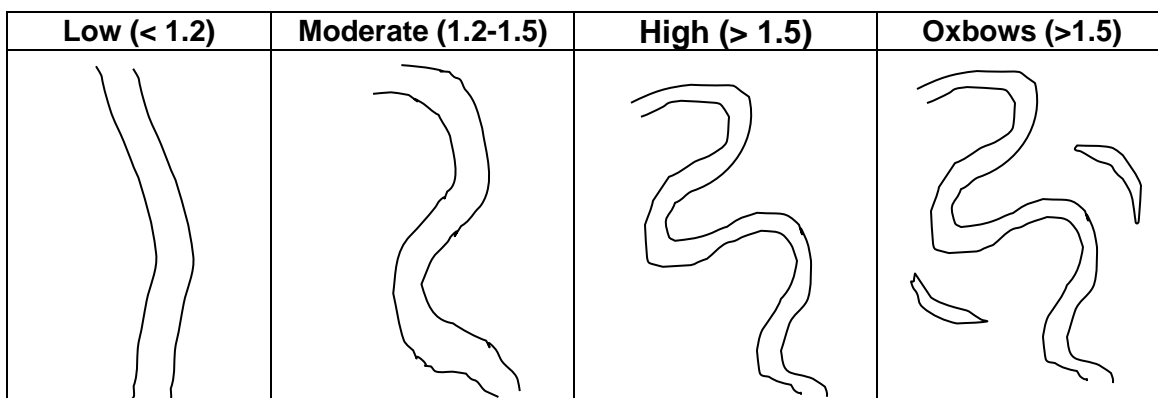


Figure 3.7.6 Diagram of different levels of sinuosity in the channel planform.

Determining Bed Forms: The Phase 2 determination of bed forms is a visual assessment. Using the descriptions provided in Table 3.7.E circle a dominant bed form on the Field Notes form for the segment (or reach). Use the dominant bed form in the Stream Type description box. If the segment exhibits more than one bed form, circle both the dominant and sub-dominant bed forms and write “dom” and “sub-dom” under them on the Field Notes form. Performing a pebble count will allow you to determine dominant bed form.

Table 3.7.E. Dominant bed form

Bed Features	Description
Cascade	Generally occur in very steep channels, narrowly confined by valley walls. Characterized by longitudinally and laterally disorganized bed materials, typically bedrock, boulders, and cobbles. Small, partial channel-spanning pools spaced < 1 channel width apart common.
Step-Pool	Often associated with steep channels, low width/depth ratios and confining valleys. Characterized by longitudinal steps formed by large particles (boulder/cobbles) organized into discrete channel-spanning accumulations that separate pools, which contain smaller sized materials. Step-pool systems exhibit pool spacing of 1 to 4 channel widths.
Plane Bed	Occur in low to high gradient and relatively straight channels, have low to high width/depth ratios, and may be either unconfined or confined by valley walls. Composed of sand to small boulder-sized particles but dominated by gravel and cobble substrates in reference stream condition. Channel lacks discrete bed features (such as pools, riffles, and point bars) and may have long stretches of featureless bed.
Riffle-Pool	Occur in moderate to low gradient and moderately sinuous channels, generally in unconfined valleys, and has well-established floodplain. Channel has undulating bed that defines a sequence of riffles, runs, pools, and bars. Pools spaced every 5 to 7 channel widths in a self-formed (alluvial) riffle-pool channel.
Dune-Ripple	Usually associated with low gradient and highly sinuous channels. Dominated by sand-sized substrates. Channel may exhibit point bars or other bedforms forced by channel geometry. Typically undulating bed does not establish distinct pools and riffles.
Bedrock	Lack a continuous alluvial bed. Some alluvial material may be temporarily stored in scour holes, or behind obstructions. Often confined by valley walls.
Braided	Multiple channel system found on steep depositional fans and deltas. Channel gradient is generally the same as the valley slope. Ongoing deposition leads to high bank erosion rates. Bed features result from the convergence/divergence process of local bed scour and sediment deposition. Unvegetated islands may shift position frequently during runoff events. High bankfull widths and very low meander (belt) widths.

Bed Sediment Composition: Using a pebble count methodology, record the percentage of each of the sediment size classes in the stream segment (or reach). See Table 3.7.F. for the classification and description.

Table 3.7.F. Bed Sediment Composition classification and description.

Size Class	Millimeters	Inches	Relative Size
1-Bedrock	> 4096	> 160	Bigger than a Volkswagen Bug
2-Boulder	256 – 4096	10.1 - 160	Basketball to Volkswagen Bug
3-Cobble	64 – 256	2.5 - 10.1	Tennis ball to basketball
4-Coarse Gravel	16 – 64	0.63 – 2.5	Marble to tennis ball
4-Fine Gravel	2-16	0.08 – 0.63	Peppercorn to marble
5-Sand	< 2.00	< 0.08	Smaller than a peppercorn
6-Silt	<.062	<.002	Smaller than sand

Determining Dominant adjustment processes:

This can be a visual assessment of the current dominant adjustment process based on visual observations of degree of incision, depositional features, presence and location of erosion, and other channel adjustment characteristics. More than one adjustment process may be active at a given time (for example, both widening and planform adjustment). See Table 3.7.G for the classification and description.

Table 3.7.G. Dominant Adjustment processes classification and description.

Process	Description
Degradation	Channel is actively incising, or has incised historically but has not progressed to later stages of channel evolution (potentially due to channel armoring). Nick points may be present; bed features may be eroded.
Aggradation	Channel is actively aggrading. Presence of depositional features such as mid channel bars, diagonal bars, side bars, and steep riffles.
Widening	Channel is actively widening. Presence of bank erosion and low w/d ratio. Bed features may be sedimented.
Planform	Channel is moving laterally. Evidence of active flood chutes, avulsions, erosion on outside bends of meanders.

Geomorphic condition: This can be a visual estimate of the streams degree of departure from reference equilibrium condition. To determine a more value based condition, an observer can use the [Rapid Geomorphic Assessment Forms](#). See Table 3.7.H. for the classification and description.

Table 3.7.H. Stream Condition classification and description.

Stream Condition	Description
Poor	stream reach in <i>poor</i> condition that is experiencing <i>extreme</i> adjustment outside the expected range of natural variability for the reference stream type; likely exhibiting a new stream type; and is expected to continue to adjust, either evolving back to the historic reference stream type or to a new stream type consistent with watershed inputs and boundary conditions
Fair	A stream reach in <i>fair</i> condition that has experienced <i>major change</i> in channel form and fluvial processes outside the expected range of natural variability; and may be poised for additional adjustment with future flooding or changes in watershed inputs that could change the stream type
Good	A stream reach in <i>good</i> condition that is in dynamic equilibrium which may involve localized, <i>minimal change</i> to its shape or location while maintaining the fluvial processes and functions of its watershed over time and within the range of natural variability.
Reference	A stream reach in <i>reference</i> condition that is in dynamic equilibrium which may involve localized, <i>insignificant change</i> to its shape or location while maintaining the fluvial processes and functions of its watershed over time and within the range of natural variability.

Stream Sensitivity: Sensitivity refers to the likelihood that a stream will respond to a watershed or local disturbance or stressor. With the help of table below, use the existing stream type and the stream condition to evaluate the sensitivity of your segment or reach. If the existing stream type represents a departure from a reference or modified reference stream type then you will use the far right-hand column of table.

Table 3.7.I Stream Sensitivity based on stream type and condition.

Existing Geomorphic Stream Type	Sensitivity		
	Reference or Good Condition	Fair-Poor Condition in Major Adjustment	Poor Condition, Represents a Stream Type Departure
A1, A2, B1, B2	Very Low	Very Low	Low
C1, C2	Very Low	Low	Moderate
G1, G2	Low	Moderate	High
F1, F2	Low	Moderate	High
B3, B4, B5	Moderate	High	High
B3c, C3, E3	Moderate	High	High
C4, C5, B4c, B5c	High	Very High	Very High

A3, A4, A5, G3, F3	High	Very High	Extreme
G4, G5, F4, F5	Very High	Very High	Extreme
D3, D4, D5	Extreme	Extreme	Extreme
C6, E4, E5, E6	High	Extreme	Extreme

Channel Evolution Sequence (Figure 3.7.7): This can be a visual assessment of the current dominant adjustment process based on visual observations of degree of incision, depositional features, presence and location of erosion, and other channel adjustment characteristics.

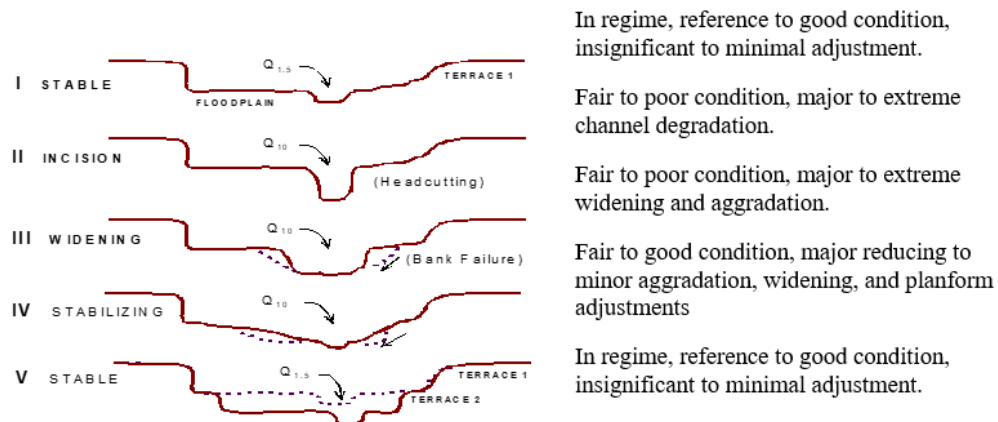


Figure 3.7.7: Five Stages of Channel Evolution (Schumm, 1977 and 1984), the channel condition and adjustment processes often observed during each stage.

Data Collection and Analysis – Existing vs. Reference:

Table 3.7.J. serves as a simple field form and first-cut analysis of data to evaluate the degree to which a segment has departed from equilibrium condition. Some of the “expected” values could come from (or be inferred from) Phase 1 SGA when available and would be useful to populate prior to fieldwork. The adjustment process column would allow you to designate an adjustment process that would explain any differences between existing and reference conditions and help inform the overall evaluation of channel condition.

Table 3.7.J. Existing vs. Reference Condition Summary

Existing vs. Reference Conditions			
Parameter	Existing	Reference	Adjustment Process
Confinement			
Bankfull width			
W/D ratio			
Entrenchment			
Stream type			
Incision			
Bed form			

Dominant substrate size			
Evaluation of Current Condition			
Geomorphic Condition			
CEM Stage/Model			
Sensitivity			
Process Narrative			

Field methods used by the Rivers Program pertaining to Stream Geomorphic Assessment are described in the Vermont Stream Geomorphic Assessment [Phase 2 Handbook: Rapid Stream Assessment](#) (2009) and [Phase 3 Handbook: Survey Assessment](#) (2009).

3.8 Physical Habitat Characterization - Lentic

Physical habitat characteristics are observed and recorded to assess both riparian and littoral habitat condition. The Lake Assessment Program uses these data to characterize the condition of the lakeshore and shallow water habitat and the impacts of shoreline development and water level manipulations. Two different methods are used to assess lentic nearshore habitat. Vermont has adopted an enhanced version of the National Lake Assessment (NLA) methods for measuring physical habitat (PHAB) as part of its Next Generation Lake Assessment Program. Methods are described in the [US EPA National Lake Assessment Field Operations Manual](#). Vermont also uses Littoral Habitat Assessments (LHA). Currently, all measurements are made on paper forms (See Lentic Physical Habitat Assessment Form in Figure A.5), except during the NLA when PHAB measurements are made using the NLA electronic field forms on an iPad or iPhone.

3.8.1 Enhanced National Lake Assessment Physical Habitat

Site Information

- Lake Name
- Site Name, A through J
- Date of sampling
- Crew Names
- Site Type: Point, Cove, Shore, Inlet, Other
- Site Developed w/in 100m? Yes or No
- Latitude and Longitude
- Dominant Habitat Type: Rocky, Macrophyte, Muddy or Sandy Littoral
- GPS Projection
- Snorkeler, Macroinvertebrate Collector, Buffer Measured By and PHAB Observer
- Photos of site using cell phone or tablet camera



National Lake Assessment Riparian Observations

- Follows protocols described in the [US EPA National Lake Assessment Field Operations Manual](#).
- Makes observations of the subset of PHAB parameters from the NLA that are used in the Northern Appalachian Ecoregions PHAB metrics, so it is a subset of the NLA observations.
- Observations are made of the riparian area from the observation station (Figure 3.8.1).
- Following the NLA protocols, percent cover of the canopy, understory, woody shrubs and saplings, non-woody shrubs and saplings, standing water or inundated vegetation and bare ground are made. Sum of cover can exceed 100%.

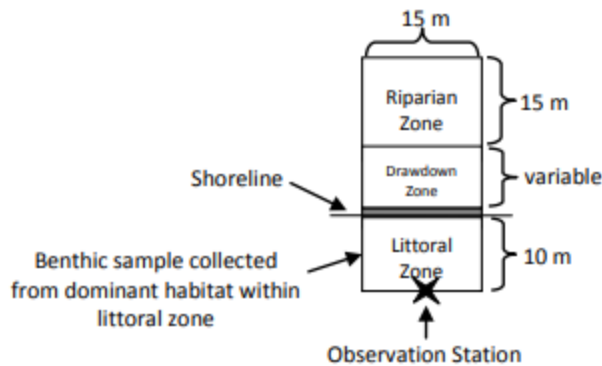


Figure 3.8.1. NLA diagram showing where observations of the riparian and littoral zone are made.

National Lake Assessment Lakeshore Disturbance & Lakes Score Card Observations

- Following the NLA protocols, presence of human disturbance inside (score 1) or outside (score 0.5) the Riparian Zone plot is noted. Score is zero if human influence category is not present. Human influence categories are buildings, park facilities/Man-made beach, commercial, docks/boats, row crops, pasture/range/hay field, orchard, lawn, walls/dikes/revetments, landfill/trash, roads/railroads and powerlines.

National Lake Assessment Shallow Water Habitat Observations

- Following the NLA protocols, percent cover of submergent, emergent and floating vegetation in the littoral zone plot are recorded.
- Enhancement to NLA, dominant 3 macrophyte species are noted.
- Following the NLA protocols, fish cover is estimated in terms of percent cover of aquatic & inundated herbaceous vegetation (this value should equal the sum of the % cover of submergent, emergent and floating leaved plants), percent cover of small and large woody debris, inundated live trees, overhanging vegetation within 1m of the surface, ledges or sharp dropoff, boulders, human structures.

Enhancements to the National Lake Assessment Observations

- Immediate shore type as viewed from the observation station for the 15m wide shoreline. Values must sum to 100% and include tree stratum >5m, High Shrub 1.5-5m, low shrub 0.1-1.5m, and ground cover. If ground cover type is noted if bare, sandy beach, or lawn.
- It is noted whether site photo was taken from the observation station, this serves as reminder to take the photo.
- The percent sand cover at 0.5m depth contour along 15m of shore site is noted.
- The percent embeddedness is estimated by picking up 3 cobbles, woody debris, gravel located along the 0.5m depth contour along the shore. If no suitable material can be found to estimate embeddedness, then it is recorded as not applicable. Embeddedness is estimated as % of cobble, woody debris or gravel that is buried in the littoral sediments.
- A densiometer reading is taken 1m from the waterline in the center of the 15m shoreline. The densiometer is modified for looking at one shore by applying a V of tape as shown in the below Figure 3.8.2. This leaves a maximum of 17 intercepts. Holding the densiometer 1' above the water, while keeping it level as indicated by the bubble in the lower right and while holding it so that the top of your head is just

visible at the bottom of the V, the number of intercepts which are shaded are counted.

- The percent cover of invasive species observed in the littoral or riparian zones are noted.

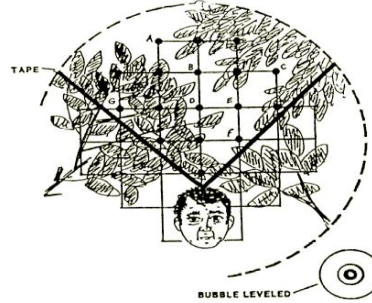


Figure 3.8.2 *Spherical Convex Densiometer modified for measurements of shoreline vegetation with 17 intercepts in V used.*

3.8.2 Littoral Habitat Assessment

Site Information

- Using data sheets printed on waterproof paper, attached to a clipboard with binder clips and using a No. 2 pencil attached with long rubber bands, all observations are made while snorkeling.
- Lake Name
- Date
- Site Number: Number plus D if Developed, U if undeveloped or B if buffered developed
- Site Type: Point, Cover, Shore, Inlet or Other
- Dominant Habitat Type of 0.5m depth contour: Rocky, Macrophyte, Muddy, Sandy or Other
- If site is developed, note if recent or established
- Crew: Terrestrial sampler, plant snorkeler, woody debris snorkeler.
- Latitude and Longitude

Site Setup

At each site, 10 m transects are established parallel to the shore at 0.5m, 1.0m and 2.0m depths (Figure 3.8.3). Depth is used instead of distance from the waterline in an attempt to capture the full diversity of plant species present in a lake. Distance from the shore varies with slope and is recorded at each site. Each transect is broken into two 1 m by 5 m plots.

- A yellow floating line with pink flagging denoting the ends of each 5m plot and bright green flagging at every meter within each plot is floated along the 0.5m depth contour. The line is anchored at each end with old railroad spikes.
- Two buoys with 1m of line attached to a dive weight are set at each end of the 1m depth contour transect.
- Two buoys with 2m of line attached to a dive weight are set at each end of the 2m depth contour transect.
- Each snorkeler uses a 1m long CPVC wand with a line in the middle and uses this as they snorkel along the 5m depth contour as a reference for the 1m by 5m plot. The snorkeler uses the bright flagging on the transect line for orientation and to help estimate % plant cover.

- Another CPVC pole that is 2.25m long is marked with 1m and 2m increments and is used to demark the center of the 2m transect that separates plots D1 and D2 (Figure 3.8.3).

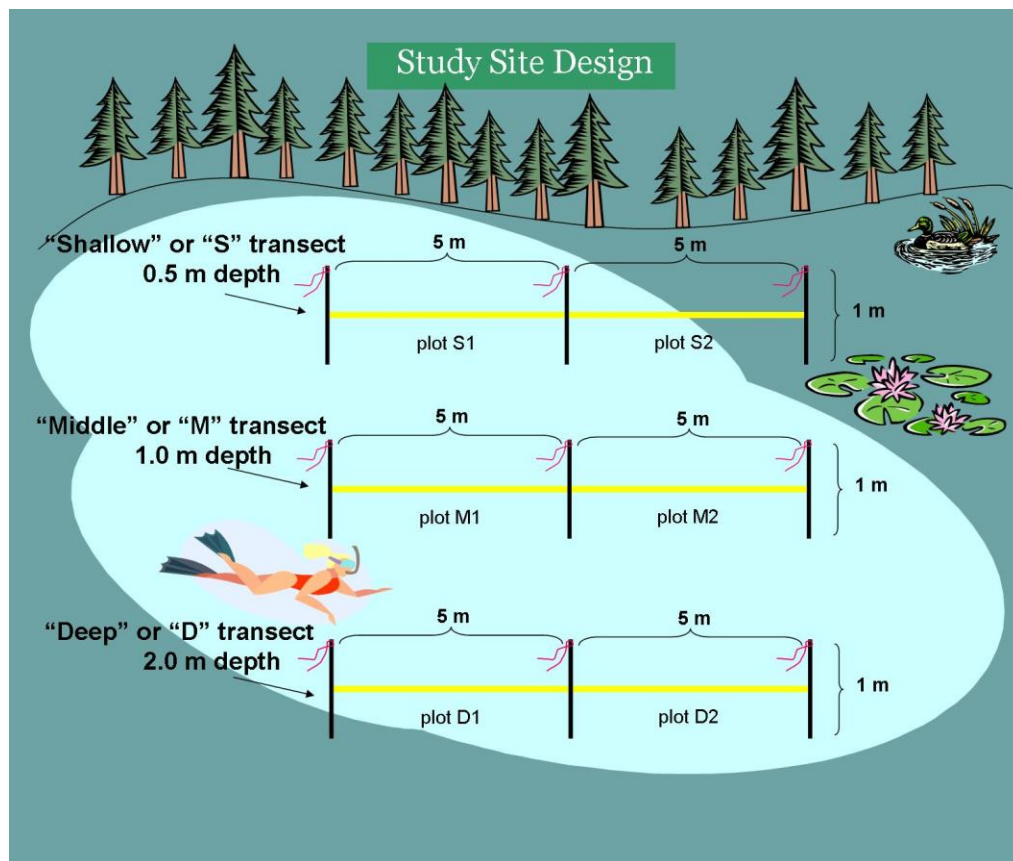


Figure 3.8.3 *Littoral Habitat Site Layout*

Littoral Habitat Assessment (LHA) Riparian Vegetation % Cover (high water mark to 10m inland)

- The riparian observations are made from the middle of the D2 transect while treading water. This is where the site photo is taken with a waterproof camera or iPhone in waterproof case with wrist float.
- Immediate shore type for the 10m wide shoreline. Values must sum to 100% and include tree stratum >5m, high shrub 1.5-5m, low shrub 0.1-1.5m, and ground cover. Ground cover type is noted if bare, sandy beach, or lawn.
- The dominant ground cover of the riparian area from the water line to 10m inland is noted (e.g., duff, lawn, ferns, etc.).

LHA Littoral Site Observations and Measurements

- Shading is measured with a densiometer at 1m and 5m from shore in the midpoint of the transect. Spherical Convex Densiometer modified for measurements of shoreline vegetation with 17 intercepts in V used (Figure 3.8.2)
- Large woody habitat (>10cm diameter) counts are made for the entire site by snorkeling with a 1 m long CPVC wand with 4cm and 10cm markings on each end. The 10cm marking is used by the snorkeler to verify diameter.

- The 2.5m CPVC pole used to demark the boundary between the deep transect plots is used to measure the distance from the ends of the 2m transect to shore. If available and the distance is far, a rangefinder is used instead. Note whether a pole or rangefinder is used. The distance (m) from the buoy located at the end of plot D1 is recorded as 2A and the distance (m) from the buoy located at the end of plot D2 is recorded as 2C.
- Exuviae of odonate nymphs are collected along the 1m wide immediate shore parallel to the 10m transects. If sampling by snorkeling occurred before known flight times for most odonates, then return visits are made to resample for odonate exuviae at the sites. Exuviae at each site are put into 50ml test tubes. Total counts of exuviae at each site are recorded.
- It is noted if macroinvertebrates were sampled. See Section 6.5 for Lentic LHA macroinvertebrate collection methods.

LHA Littoral Plot Observations

- Percent cover for fine woody habitat (<4cm diameter), medium woody habitat (4-10cm diameter), deciduous leaf litter, aquatic plant species, periphyton and aufwuchs are estimated by snorkeler.
- Thickness of both periphyton and aufwuchs is noted as 0, slimy, draw line, 0.5-1mm, 1-5mm, 5-20mm, or >20mm.
- Estimation of embeddedness is made in each plot. Plot sediment is defined in proportions of 9 types (sand, muck, floc, gravel, cobble, boulder, silt, ledge, woody substrate).
- Percent cover of individual macrophyte species is estimated in each plot.
- Presence of fish, mussels, crayfish or snails is noted for each plot.

3.8.3 Continuous Dissolved Oxygen Sensors

Follows Regional Lake Monitoring protocols currently in draft form with Tetra Tech and EPA.

3.8.4 Continuous Water Level Pressure Sensors

Follows Regional Lake Monitoring protocols currently in draft form with Tetra Tech and EPA.

4 In Situ Water Chemistry Measurements

4.1 Field pH

For low pH solutions (<pH 6), use a calomel electrode. For high pH solutions (>pH 10), use a “low sodium error” electrode. For solutions closer to neutral, a standard pH probe is adequate.

- a. Storage:
 - i. Never store a pH probe in distilled water.
 - ii. For short period of times (<1 week), pH electrodes should be stored with their fill hole covered and the ceramic junction submerged in electrode- storage solution.
 - iii. For non-refillable probes, cover the ceramic junction with a protective cap filled with electrode storage solution.
 - iv. For longer period of times, the sensing bulb of the pH probe should be covered with a protective cap filled with storage solution and the fill hole covered.
- b. Maintenance:
 - i. The solution level in refillable electrodes should be checked frequently and replenished as needed with the appropriate electrolyte.
 - ii. Gel filled reference electrodes leak the slowest but are not refillable and will generally run out of reference solution in about a year. These should be replaced every 6 months.
 - iii. If crystals form inside a refillable electrode, or if the pH probe starts to drift or respond slowly, drain and replace the filling solution.
 - iv. Avoid wiping the bulb of a pH electrode. Electric static may occur and cause polarization.

The Hanna Waterproof Portable pH Meter Model 9025 meter is primarily used by the Vermont Long-Term Monitoring Program (VLTm) for acid sensitive lakes. It is equipped with a calomel reference pH electrode which more accurately measures low ionic strength (low pH) solutions.

Equipment:

- pH meter
- pH electrode
- ATC Temperature probe
- Kimwipes
- Distilled water
- Sample cup (glass or plastic beaker)
- pH buffers (4, 7 and/or 10)

Calibration Procedure:

1. When calibrating a pH meter, use fresh certified standard pH buffers. The two buffers selected should be no more than 3 pH units apart and should bracket the expected pH value. Use pH 4, pH 7 and/or pH 10 buffers daily. Ensure that the electrode filling solution is above the ceramic junction and at least one inch above the sample level when immersed. If not, fill electrode with 4m KCl electrode filling solution. Rinse out and refill the electrode if the solution has crystallized.
2. Attach the pH electrode and ATC temperature probe to the BNC connector and turn meter on.

3. Remove protective cap from pH electrode tip. Rinse pH electrode and ATC probe with distilled water and pat dry with a Kimwipe.
4. Put pH electrode and ATC probe in pH buffer 7 buffer solution, stir gently and press "CAL" button. Display will flash "CAL" and "NOT READY". The bottom of the screen displays the buffer the meter is looking for. The default value for the first buffer is 4. Use the up and down arrow keys to bring the value to 7.01. When reading has stabilized, the meter's screen will read "CAL, READY", and "CON" will be flashing. Press "CFM" to confirm the first calibrant.
5. Rinse pH electrode and ATC probe with distilled water and pat dry with a Kimwipe.
6. Put pH electrode and ATC probe in pH 4 buffer solution and stir gently. Make sure the lower right corner display reads "BUF 2 4.01 pH".
7. Display will flash "CAL" and "NOT READY" until the reading for the second buffer has stabilized. When reading has stabilized, the meter's screen will read "CAL, READY", and "CON" will be flashing. Press "CFM" to confirm the second calibrant. If the second buffer is pH 10, use the up and down arrow to adjust the value for the second buffer. The meter is now calibrated and ready to read samples.

Measurement Procedure

1. Rinse pH electrode and ATC probe with distilled water and pat dry with a Kimwipe. Rinse sample cup with distilled water, then rinse sample cup with sample water.
2. Dip pH and temperature probes into sample water and gently stir. Wait for reading to stabilize. Record reading.
3. Turn off meter. Replace cap on probe filled with buffer solution to prevent wick from drying out. Cover filling hole on pH probe with protective sleeve.

4.2 Field Conductivity

Field conductivity is measured using a conductance meter and a cell (probe). The meter and cell should be calibrated frequently using potassium chloride standards of known conductance. WSMD currently uses YSI conductivity meters, Hydrolab multi-probes and PC Testers. Use calibration standards with values above and below the expected range of the samples. Loggers are also now widely available for taking 'continuous' conductivity measurements at short time intervals, which can help capture variability during runoff events or low flow periods.



4.2.1 Hanna HI 9033 Portable Waterproof Conductivity Meter

Calibration procedure:

1. Unlike pH meters, a conductivity meter need not be calibrated daily. Turning off the instrument will not affect the meter's accuracy. A standard should be used at least weekly to verify the accuracy of results. Use a calibrant that is close to the conductivity of the waters being measured. The calibration solution should be at 25°C when measured.
2. Fill a clean beaker with calibrant. Make sure probe is connected to the meter by pushing in the plug and tightening the threaded pin. Rinse conductivity probe with distilled water and pat dry with a Kimwipe. Rinse conductivity probe with sample water. Dip probe into calibrant. Make sure that the sample water is above the air release holes on the probe.
3. Turn meter on by pressing the "ON/OFF" button and select the appropriate range (e.g. 199.9 $\mu\text{S}/\text{cm}$). Tap the conductivity probe on the bottom of the beaker in order to release any air bubbles caught inside the probe.
4. Allow reading to stabilize.
5. Use a flathead screwdriver to adjust the calibration trimmer until the meter reads the correct conductivity value. All measurement will be temperature compensated to 25°C.

Measurement procedure:

1. Turn on the meter. Rinse the cell with distilled water and pat dry with a Kimwipe.
2. Place cell into waterbody at the desired depth, making sure the water comes above the cell holes. Drift may occur when placed directly in flowing water. With cell in sample bottle, select one of the four buttons with the expected range of, i.e. 199.9 μS , 1999 μS , 19.99 mS or 199.9 mS. Wait for the reading to stabilize and recorded the value (typically within one minute).
3. Remove cell from water, and rinse with distilled water. Pat dry cell and return meter and cell to carrying case.

4.2.2 Onset HOBO U24 Conductivity Data Logger

WSMD periodically deploys conductivity data loggers in freshwater streams to collect a time-series of specific conductivity over an extended period. Onset's HOBO U24 Conductivity

Loggers are currently relied on for these applications. Much of the operation and maintenance methods follow those specified in the latest product user manual (Onset, 2019) and with the aid of the Conductivity Assistant in Onset's HOBOWare software program. Use of continuous data loggers with co-located chloride grab samples over a range of conditions allow for the development of conductivity-chloride rating curves and time-series of in-stream chloride concentrations. Where concurrent streamflow data exist, deriving chloride loading over time is possible.

1. **Logger calibration:** Calibration adjustments are not made to the conductivity sensor itself, rather data are corrected within HOBOWare during post-processing with the use of independent measurements of actual conductivity via thermometer and a calibrated conductivity meter. For open-channel surface waters, the preferred method involves collecting in-stream temperature and conductivity readings at the logger location for both the beginning and end of each deployment period. The corresponding conductivity data offloaded from the U24 logger are then corrected to the reference measurements within HOBOWare. See user manual for additional details (Onset, 2019).
2. **Lotic temporary deployment methods:** Deployment methods for conductivity loggers are similar to those of water level and temperature loggers. Conductivity loggers are placed in PVC stilling wells or protective housings that are perforated to allow full circulation of ambient stream water around the sensor. The PVC housing is securely affixed to either a piece of rebar or fence post driven into the stream bed, epoxied to the side of a boulder or submerged concrete face, or a board anchored into boulder, bedrock ledge or concrete of bridges and culverts. See Sections 3.1 and 3.4 for additional information on in-stream deployment techniques.
3. **Data QAQC:** QAQC procedures follow those recommended in the U24 logger user manual (Onset, 2019), including calibration methods. General site visit and deployment equipment maintenance are also like those described in sections 3.1 and 3.4 for continuous temperature and water level deployments. This includes proper documentation of deployment and removal times, trimming errant data points impacted by off-loading activity and regular site visits to clean biofouling, addressing sedimentation and maintenance to prevent hardware failure and equipment loss. Proper calibration and maintenance of portable conductivity meters and multi-sondes (Sections 4.2 and 4.3) are important to QAQC procedures for conductivity loggers.

4.3 Measurements Using a Multiple Parameter Probe

WSMD field staff currently use Hydrolab DS5, MS5 and Compact MS5 multiprobes with Hydrolab Surveyor 4a to measure water quality parameters in-situ in various waterbody types. These multiprobes have two components: a sonde containing multiple sensors which is submerged in a waterbody and a surveyor unit which reads and/or stores the data. The calibration and maintenance procedures described here are vital to equipment longevity and accurate readings. More detailed technical information regarding multiprobe and surveyor calibration, maintenance, and usage can be found in the [Hydrolab Water Quality Multiprobe User Manual](#) (Hach Company, 2006) and the Surveyor 4a User Manual (Hach Company, 2004).

Equipment:

- Hydrolab multiprobe
- Hydrolab surveyor
- Calibration cap
- Cable
- Calibrants
- Deionized (DI) water for LDO calibration
- Tap water for storage
- Protective case for storage and transport

Hydrolab multiprobe sensors:

- Specific conductivity ($\mu\text{S}/\text{cm}$)
- Turbidity (NTU)*
- pH
- Dissolved Oxygen (mg/L and % saturation)
- Temperature (degrees Celsius)
- Chlorophyll-a*
- Depth (m)



*Not all probes are equipped with these sensors.

4.3.1 Calibration Procedures

Multiprobe sensors for pH, specific conductivity, and turbidity (if applicable) are calibrated in the laboratory once per week during WSMD field seasons and results are recorded on calibration record sheets (Figure 4.3.B). A two-point calibration is used for specific conductivity, pH and turbidity as outlined in Table 4.3.A. Standard calibrants for pH, specific conductivity, and turbidity are maintained at the Watershed Management Division Laboratory at VAEL. Each calibration procedure is described below.

Table 4.3.A Two-point calibration for select multiprobe parameters.

Parameter	Calibration point 1	Calibration point 2
Specific conductivity	Air (SpC = 0 $\mu\text{S}/\text{cm}$)	84 or 500 $\mu\text{S}/\text{cm}$
pH	7	4 or 10
Turbidity	DI water (Turbidity = 0.3 NTU)	10, 25, or 40 NTU

4.3.1.1 Specific conductivity, pH, and turbidity

1. Prior to laboratory calibration, review the MSDS forms for each calibrant.
2. Wear safety glasses and gloves.
3. Connect the multiprobe to the surveyor using the appropriate cable. Line up the dots and do not twist. Turn the surveyor on (O||).
4. With calibration cup in place, rinse sensors 3 times with DI water.
5. Fill the calibration cup with 1 inch of calibrant standard for calibration point 1 to rinse sensors and calibration cup. Expired or “used one time” calibrant may be used for rinsing.
6. Repeat for a total of 3 rinses. Dispose of used calibrant in a labeled waste container, preferably a previously used container for the same calibrant.
7. Fill the calibration cup so the sensor is covered with fresh calibrant. Wait 30-60 seconds for the readings to settle and record the pre-calibration value on the calibration sheet.
8. On the Surveyor, press ‘Setup/Cal’, ‘Calibrate’, ‘Sonde’. Use the arrows to navigate to the parameter and press ‘Select’.
9. Enter the calibrant standard value for calibration point 1 and press ‘Done’.
10. Wait 30-60 seconds for the readings to settle and record the post-calibration value on the calibration sheet.
11. Note any issues on the calibration sheet.
12. Repeat steps 4-11 for the second calibration point.
13. When disconnecting cable, carefully pull straight off – DO NOT TWIST.

Notes on laboratory calibration:

- When calibrating the turbidity sensor, the user will need to specify calibration point 1 and 2 in the surveyor prior to entering calibrant standard values. Turbidity calibrants should be handled with caution and should be gently inverted and swirled to homogenize prior to use. Do not shake or bubbles will form, causing an inaccurate calibration. ‘Set up’, ‘Calibrate’, ‘Sonde’, ‘Clean’, ‘Revolutions’, ‘1’ can be used to clean the turbidity sensor.
- The air spigot in the Watershed Management Division Laboratory or the exhaust hose on a vacuum pump can be used to quickly dry the specific conductivity sensor for calibration point 1 (air).
- Issues with calibration should be recorded on the calibration sheet and use of the sensor in question should be discontinued until repairs are made.

4.3.1.2 Dissolved Oxygen

Dissolved oxygen (DO) measurement is dependent upon barometric pressure and must be calibrated at the field site immediately prior to data collection. Measure the barometric pressure on-site in mm Hg using an iPhone app or the surveyor. Calibrate DO according to the type of sensor used following the steps below. The circulator should be turned off during calibration and turned back on again after calibration (lakes only). To turn circulator on or off select ‘Setup/Cal’ on Surveyor, ‘Setup’, ‘Sonde’, Select ‘Circltr: Off/On’, Select ‘0’ for Off and ‘1’ for On, ‘Done’, Hit any key, Hit ‘Go Back’ until Go Back is no longer an option. WSMD Hydrolabs are equipped with either a Clark Cell membrane or LDO sensor for dissolved oxygen measurements.

1. Clark Cell Membrane

- a. Make sure there are no bubbles under the membrane. If bubbles are present the membrane must be replaced. Once the DO membrane is replaced, wait overnight before attempting to calibrate again to allow sensor to equilibrate.
- b. Cover the sensors with ambient water to just below the 'O' ring of the DO sensor membrane.
- c. Keep sensors pointed up.
- d. Gently rest cap on top, do not screw on (this keeps temperature from fluctuating).
- e. Select 'Setup/Cal' on Surveyor, 'Calibrate', 'Sonde', 'DO%', then enter BP in mmHg, 'Done', Hit any key, 'Go Back'.
- f. When disconnecting cable, carefully pull straight off – DO NOT TWIST.

2. LDO Sensor (black capped sensor)

- a. Make sure no more than 1/3 of black color removed from cap, otherwise change cap.
- b. A 0.5 degree C change in temperature while calibrating will make values generated untrustworthy. Make sure water used to calibrate is at air temperature before starting calibration. Keep ~600ml of DI water in 1L TSS bottle with the DS5 unit to achieve this.
- c. Shake 1L TSS bottle with ~600ml of DI for **50** seconds. Ambient water may be used if its conductivity is less than 500 $\mu\text{S}/\text{cm}$.
- d. With sensors pointing upward, fill calibration cup to bottom thread at top of cup with oxygenated water from shaken bottle. Invert cap and balance on top of cup.
- e. Make sure temperature is stable (<0.5 degree C change).
- f. Select 'Setup/Cal' on Surveyor, 'Calibrate', 'Sonde', 'LDO%', then enter barometric pressure in mmHg, 'Done', Hit any key, 'Go Back'.
- g. When disconnecting cable, carefully pull straight off – DO NOT TWIST.

4.3.2 General Maintenance

- Always store multiprobes with calibration cup on and 1-2 inches of ambient water or tap water during storage and transport. Never store dry or filled with DI water.
- Recharge the surveyor at the end of a field day:
 - On last run turn off Timed shutdown (Setup/Cal, Setup, Timeout: Shutdown, 0 to disable)
 - Leave unit on to discharge battery completely (usually lasts 24 hrs total)
 - Recharge 3.5 hrs (plug in and turn on)
 - Turn Timeout on to save charge (Setup/Cal, Setup, Timeout: Shutdown, 5)
- Upon returning to the lab
 - Empty calibration cup of ambient water, rinse 3 times with tap water and store with 1-2 inches of tap water overnight.
 - The surveyor, Hydrolab case, and everything except the sensors should be allowed to dry.
 - Plug in and turn on the surveyor to recharge the battery.
- Multiprobes with the Clark Cell DO membranes need to be replaced when they become ripped or if bubbles form. When this happens, the sensor can no longer read DO and

field calibration will fail. Refer to the [Hydrolab User Manual](#) and follow the steps [in this video](#) to replace the membrane.

- Refer to the Hach LDO Sensor Instruction Sheet for instructions on LDO cap maintenance and replacement.
- For long-term storage, use 1-2 inches of 500 $\mu\text{S}/\text{cm}$ specific conductivity calibrant or Hydrolab storage solution.
- Once per month or as sensors foul, do the following:
 - Change DO membrane & electrolyte, make sure there are no bubbles under the membrane
 - Change pH electrolyte
 - Clean glass pH probe with methanol
 - Clean conductivity port, removing O-rings and buffing prongs
 - Clean Chlorophyll a glass surfaces with lens paper

Hydrolab Calibration & Service Record, 2017 Version						
Hydrolab Unit (circle one):		Big Mama	Oreo	Old White	Turb-0	Baby Bass
Date:		Calibrator:				
Parameter	Standard	Pre-Calibration Reading	Post-Calibration Reading	Comments		
pH	7.00					
	4 -or- 10					
Conductivity	0.0 (air)					
Chl-a	0.0 (DI)					
Turbidity	0.3 (DI)					
Servicing Notes:					Comments	
Clark Cell DO Membrane Changed?	Yes	No				
DO Electrolyte Changed?	Yes	No				
LDO Cap Changed?	Yes	No				
pH Reference Electrolyte Changed?	Yes	No				
Salt tablet added to pH electrolyte?	Yes	No				
Wiper replaced on Turbidity Sensor?	Yes	No				
All sensors scrubbed gently with toothbrush and soapy water?	Yes	No				
Clean conductivity sensor (Q tip for new, sand paper and DI for old)	Yes	No				
Describe any other service	Yes	No				

Figure 4.3.B Hydrolab calibration & service record

4.3.3 Field operations in lotic environments

- Store sonde in protective case with 1-2 inches of tap water in calibration cup in between sites. Shut off surveyor in between sites to conserve battery.
- Upon arrival to a site, connect the sonde to the surveyor by lining up the dots on the cable and pushing straight on. Do not twist. Turn surveyor on.
- Empty tap water in calibration cup into storage bottle on site. Calibrate dissolved oxygen at each site following instructions in 4.3.1.2.

- d. After dissolved oxygen is successfully calibrated, remove calibration cup and set aside. Screw on weighted probe protector and gently place sonde in flowing water.
- e. Wait 1-2 minutes for pH readings to stabilize. Record values.
- f. After data are recorded, remove sonde from water, take off probe protector and put back calibration cup with 1-2 inches of tap water.
- g. Unplug and turn off surveyor and return components to travel case.

4.3.4 Field operations in lentic environments

Calibrate depth at each lake:

- a. Setup/Cal
- b. Calibrate
- c. Sonde
- d. Select 'Dep100: meters'
- e. Hang Hydrolab sensors to 1 m depth
- f. Select 1.0
- g. Done
- h. Hit any key
- i. Hit Go Back until Go Back is no longer a selection

Annotate files at each different site:

- a. Files
- b. Svr4a
- c. Annotate
- d. Select 'Lake Assessment'
- e. Enter name of site
- f. Done
- g. Hit any key
- h. Hit Go Back until Go Back is no longer a selection

Record readings

- a. Hang or lie Hydrolab in stream or lake
- b. Wait until temperature equilibrates
- c. Hit 'Store'
- d. Select 'Lake Assessment'
- e. Wait until logging active note disappears
- f. Record on backup data sheet
- g. If in lake, drop at 1 m intervals with last reading 1 m above bottom
- h. At each depth equilibrate DO & hit 'Store'
- i. If in lake, pull up to 1 m, equilibrate and hit 'Store' to check pH against 1st reading

Review files to see what logged in

- a. Files
- b. Svr4a
- c. Review
- d. Select Lake Assessment'
- e. Choose to review from beginning or by date and time
- f. Done
- g. Hit 'Go Back' until Go Back is no longer a selection

4.3.5 Transferring data from Surveyor to Excel

Exporting data from surveyor:

- a. Hook up serial port cable to computer and Surveyor
- b. Open up C:\Program Files\Accessories\Hydrolab.ht
- c. Turn on Surveyor
- d. Send file from Surveyor: Hit 'Files' key, Choose 'Transmit', 'Bass Remap' or 'Lake Assessment', 'SS-Importable'
- e. On Computer: Go to 'Transfer' tab and choose 'Receive File'
- f. At 'Place received file in the following folder' prompt, enter folder name
- g. Use receiving protocol: 'Xmodem'
- h. Hit 'Receive'
- i. Enter file name. Use project and date download and .txt extension (i.e., Remap06-10-20.txt)
- j. Close program

Transferring data to excel:

- a. Open Excel
- b. Open .txt file
- c. Choose boxes for Delimiters Tab and Commas
- d. Skip any blank columns or data don't want or don't trust (faulty sensor, salinity).
- e. Finish (should see data imported now)
- f. Check for outliers, redundancy, etc.
- g. Save file

Clear log on Surveyor:

- a. Verify data successfully transferred to Excel file before doing this
- b. Select 'Files', 'Wipe', 'Lake Assessment', Select '1' for yes, 'Done'

5 Water Sample Collection Methods

5.1 Manual Water Sample Collection Procedures

5.1.1 Grab Samples

1. Collect surface grab samples by hand or using a dip sampler. The dip sampler can be used to obtain a more representative sample from a river or lake by collecting the sample further from shore without entering the waterbody. Rinse the dip sampler with sample water three times before collecting the sample.
2. To collect a lake grab sample, rinse sampling container three times with surface water. Then, invert the mouth of the sample container until the desired sample collection depth, keeping hands and clothing away from the mouth of the bottle. Once the container is full, bring to surface and tighten the lid securely. **DO NOT RINSE** the sample container if sampling for the following parameters: Total Phosphorus, Dissolved Phosphorus or *E. coli*.
3. To collect a stream or river grab sample, wait for any disturbed sediment to flow downstream, then face upstream when collecting the sample to avoid contamination of the sample. Rinse bottle three times with ambient water, then fill the bottle to the required level with ambient water and tighten the lid securely. **DO NOT RINSE** the sample container if sampling for the following parameters: Total Phosphorus, Dissolved Phosphorus or *E. coli*.
4. When collecting duplicates, two open sample containers should be submerged side-by-side and collected simultaneously to achieve a true duplicate sample. Field blanks are collected at the same time as sampling, except that distilled water from a lab-cleaned carboy is used in place of ambient sample water.
5. Samples requiring field filtering (e.g. dissolved concentrations) should be performed in accordance with Section 5.3.2.

5.1.2 Hose Sampling

Equipment:

- Weighted hose with attached rope marked in meters
- 5-gallon plastic bucket (acid rinsed)

Procedure:

1. Hose samples are collected to obtain vertical composite samples in lakes or very slow-moving rivers. Care must be taken to keep the open ends of the hose free from contamination by covering each end when not in use. The two ends of a garden hose can be screwed together as a method of sealing to protect the ends from contamination.
2. The hose must be weighted on one end, marked in meters, and have a rope attached to the weighted end. Determine the depth at the sample location prior to lowering the hose. Contamination of the sample by contact with the lake bottom must be avoided. The hose should be lowered to no more than one meter above the bottom.
3. Rinse the hose and the sample bucket three times prior to sample collection. Lower the rope and hose at a slow, even rate to the desired depth while keeping the hose taut and the rope loose.
4. Place two crimps in the hose by folding it over against itself just above the water surface when at the correct depth. Hold the crimped end higher than the rest of the hose, to

prevent back-wash into the back end of the hose. Slowly pull up the rope until the weighted end reaches the surface, then lift the hose to the sample container. Let the hose hang in the water while the rope is being pulled in.

5. Keeping the crimped end as high as possible, release the crimp and run the hose through both hands until all the water has emptied into the sample container. Do not let the water sample contact the weights on the end of the hose.
6. Swirl the water in the 5-gallon bucket to mix it thoroughly before filling other sample containers.

5.1.3 Kemmerer Water Sampler

Procedure:

1. Unwrap sufficient line to reach the desired sampling depth which is marked in meters.
2. Cock the Kemmerer and check to be sure it is secure. Rinse the Kemmerer three times with surface water. Check the release valve to ensure it is closed before sampling.
3. Lower the Kemmerer smoothly to the desired sampling depth, holding the messenger in hand. Some tension should be maintained on the line to ensure that the Kemmerer remains upright and cocked. A jerk on the line could trip the mechanism. If a lake profile is being taken, sample collection should begin at the surface and progress downward so that the lower depths are not disturbed prior to sample collection.
4. Drop the messenger and wait for the sound and the tug on the line that indicates the sampler has closed. If no tug is felt, the sampler may have been tripped earlier. If sampling from a distance above the water surface (such as from a bridge), a lighter messenger should be used to avoid damage to the Kemmerer.
5. Pull the Kemmerer up, maintaining tension on the line. When at the surface, hold the Kemmerer by the rope, the brass or plastic rod sticking out the top, or with a hand-held over the bottom. This will maintain tension of the Kemmerer and ensure it remains closed.
6. Push up on the spring, release pinch clamp or turn nozzle (depending on model used) to start the water flowing out the plastic tube on the bottom. Flush out at least the length of the tube or nozzle before starting to fill a sample bottle.
7. If the Kemmerer has a tube, place the tube in the sample bottle and fill the bottle to the proper depth. With a biological oxygen demand (B.O.D.) or dissolved oxygen (D.O.) bottle, the tube should touch the bottom of the bottle while filling. Allow the water to overflow the sample bottle approximately twice its volume to rid container of air bubbles.
8. With all other sample bottles, be sure to keep the tube or nozzle above the sample water to avoid contamination. Release the spring, replace pinch clamp or turn nozzle to stop the water flow when the bottle is full.
9. When finished, open the top of the Kemmerer and drain remaining water.
10. Store Kemmerer on its side, in the open position. This allows air to dry the interior and prevents mold or mildew from forming.



5.1.4 Integrated Tube Sampler

The Minnesota Pollution Control Agency 2-meter Integrated Tube Sampler method is described in the [National Lake Assessment Field Operations Manual](#) Section 5.5.1.

5.1.5 Depth Integrating Suspended Sediment Samplers

Equipment:

- DH 59 “bomb” sampler with one-pint glass milk bottle
- Plastic nozzle (if sampling deep and fast flowing water)
- DI water
- Churn Splitter

Procedure:

1. Determine need for multiple sample locations across a river to capture a representative sample
 - a. Smaller rivers (Black, Barton, Clyde) may only need one sample location if smaller width, well mixed and there is no major tributaries close to the sampling location upstream.
 - b. Larger streams such as the Winooski and Lamoille can use three sample locations if there may be difference in water quality across the river channel. These samples are combined using a churn splitter.
2. Measure depth to bottom at each sample location by lowering the DH 59 without the glass bottle in place until it hits the bottom. Raise the DH 59 sampler 0.5 meters and mark that location on rope with a knot at railing/cement wall edge to allow for sampling to 0.5 m depth.
3. Rinse sample bottle and bomb sampler inlet area with DI water 3 times.
4. Rinse sample bottle and bomb sampler with tributary water once by lowering into water column.
5. Lower the bomb sampler to water surface pointing upstream and then lower through the water column at the centroid of flow. Lower at a constant speed until the marking for that river on the rope is even with the railing/cement wall edge.
6. Pull the bomb sampler up at the same speed up through the water as it was lowered, to get a uniform sample from 0.5 m above the bottom to the top of the water column. Bubbles coming out of the sample container when it is pulled up through the surface indicate that water was collected from all parts of the column.
7. Discard sample and resample if glass bottle is full or if bottom is hit.
8. Swirl sample bottle and decant to sample bottles (and rinse sample bottles as necessary dependent on sample parameter).
9. Blank samples should be taken by rinsing the glass bottle 3 times, filling with distilled water, then decanting to sample bottles.
10. Store the bomb sampler and glass container in a box with 75% ethanol to prevent spread of aquatic invasive species between waterbodies.

Refer to the following resources for more information:

- [Lake Champlain Diagnostic-Feasibility Study Final Report \(1997\)](#)

- [USGS Field Methods for Measurement of Fluvial Sediment](#)
- [USGS Instructions for Sampling with Depth-Integrating Suspended-Sediment Samplers US D-59 and DH-76](#)
- [Churn Sample Splitter Instructions](#)

5.1.6 Automatic Sampling Devices

ISCO 2900 Portable Sampler

An ISCO Automatic sampler can be used to sample remote locations and/or for sampling a location frequently when manual sampling is inefficient or otherwise prohibitive. The sampler consists of a weather tight plastic body to protect the samples, the instrumentation, a solar panel and tubing which runs from the unit to the waterbody being sampled.



Procedure:

1. Before use, charge ISCO lead-acid battery pack.
2. Clean ISCO sample bottles, lids and collection tubing.
3. Install the ISCO sampler in a flat area and secure with stakes.
4. Program the volume and frequency of samples to be collected.
5. Refer to [Model 2900 Sampler Instruction Manual](#) (ISCO 1996) for more detailed directions.

5.2 Preventing Sample Contamination

5.2.1 Preventing Chemical Contamination

1. Clean sampling devices used in relatively clean waters by rinsing device with ambient waters at each sampling site prior to sample collection.
2. Clean sampling devices used in known or suspected contaminated waters routinely between samples to prevent cross contamination.
3. Treatment technique should be dictated by the analysis to be performed and type of samples collected. Generally, non-metallic samplers can be washed with tap water, followed by a thorough rinsing with distilled water. Metallic samplers and all metal parts on equipment should be washed with a non-phosphorus laboratory grade detergent and thoroughly rinsed with distilled water. Equipment used to collect samples for organic analyses should be washed with a non-phosphorus laboratory grade solution, followed by three or more rinses with organic-free water.
4. An alternative to field washing sampling devices in between samples is to bring enough devices so that at no time does the same equipment have to collect samples at more than one site.

5.2.2 Assessing and Controlling Sample Device Contamination

Most environmental sampling or analyses offers numerous opportunities for contamination. Common sources for error in environmental measurements are listed in Table 5.2.A below.

Table 5.2.A. Potential Sources of Sample Contamination

Critical Steps in the Sampling and Analytical Process	Contamination Sources
Sample Collection	<ul style="list-style-type: none"> -Equipment and apparatus -Handling (e.g., filtration, compositing and aliquot taking), preservatives (acids) -Ambient contamination -Sample containers
Sample Transport and Storage	<ul style="list-style-type: none"> -Sample containers -Cross-contamination from other samples or reagents -Sample handling -Melted ice water in sample transport cooler
Sample Preparation	<ul style="list-style-type: none"> -Glassware -Reagents -Ambient contamination -Sample handling
Sample Analysis	<ul style="list-style-type: none"> -Syringes used for sample injections -Carry-over and memory (residual from previous samples) effects -Glassware, equipment, and apparatus -Reagents (e.g., carrier gasses and eluents)

The analytical tool most used to control and/or assess sample contamination in the field and in the laboratory are blanks. They will contain negligible amounts of the contaminants of interest and are used to estimate random errors. Blanks used to control the analytical phase are part of a laboratory's quality assurance plan, such as [VAEL Quality Systems Manual](#) (VAEL QSM). Laboratory blanks are used by analytical chemists; field blanks are used by field staff. Field blanks are used to assess contamination introduced during the collection process. Filter blanks are used to assess contamination introduced during the sample filtration process.

Procedure:

1. A field blank is generated by taking clean sampling containers to the sample collection site.
 - a. After a sample is taken, the sampling equipment is cleaned according to the standard operating procedure and rinsed with laboratory distilled water.
 - b. The final rinse is collected in the sample container(s) for subsequent analysis. Apply sample preservation procedures to the field blank.
2. Sampling Media (Trip) Blanks are used to measure contamination which occurs during transportation or collection.
 - a. VTDEC has used trip blanks samples for volatile organic chemicals.
 - b. Trip blanks determine if chlorinated and/or fluorinated volatile organic chemicals are penetrating the sample container during collection or transport.
 - c. A broader use of trip blanks involves taking an aliquot of laboratory water into the field and treating it as a sample. This includes filtration and preservation activities and allowing it to be exposed to the air.
3. Filter blanks are used to measure contamination which occurs during the sample filtration process.
 - a. After a sample is filtered, the sampling equipment is cleaned according to the standard operating procedure and rinsed with laboratory distilled water.
 - b. A new filter paper is placed in the filtering equipment, and laboratory distilled water is filtered, duplicating the ambient sample filtering protocol. Dispense

- filtered water into sample container(s) for subsequent analysis. Apply relevant sample preservation procedures to the field blank.
4. For more information, refer to Principles of Environmental Sampling, Keith, L. Ed. American Chemical Society. 1988.

5.3 Parameter-Specific Collection and Preservation Procedures

5.3.1 Sample Labels, Containers and Preservation

Most water chemistry samples collected by WSMD field staff are submitted to the [Vermont Agriculture and Environmental Laboratory](#) (VAEL) for analysis. Field staff are responsible for coordinating sample submission with VAEL staff by submitting an initial client registration form, an annual Sample Submission Plan and sample bottle order at least 2 weeks prior to sampling. After approval of the Sample Submission Plan, programs use the [WinLIMS](#) online portal to pre-log samples they plan to collect and print sample labels. Labels include the VAEL Sample ID, Customer Sample ID (typically the site name), and parameter. Labels can be printed at VAEL and in most cases, affixed to sample bottles prior to sampling. Refer to the latest version of the [VAEL Quality Systems Manual](#) when planning for water chemistry collection and analysis.

Collect all water samples in the container type and in the volume specified by parameter in the most recent [VAEL Methods](#) document. Preservation and filtering requirements, parameter-specific holding times and reporting limits are also described in this document. Sample submission instructions are subject to change and sampling staff should refer to VAEL annually for the current guidance on sample collection and submission.

In the case of spontaneous sampling (e.g., in response to a spill or other event), labels can be created and affixed to sample bottles after collection has occurred. In these instances, careful temporary labeling is imperative to ensure results are linked to the proper sampling location, date, time, and parameter (e.g. filtered vs. unfiltered). Coordinate the collection of any samples not included in the Sample Submission Plan with VAEL ahead of time.

Preserve samples that need to be kept cold (4 degrees Celsius) in refrigeration or in a cooler on ice until time of submission to VAEL. Samples that need to be preserved with acid (e.g. H₂SO₄ for nitrogen samples and HNO₃ for metals samples) can be preserved using the acidification station in the WSMD laboratory at VAEL. Samples that are to be analyzed for dissolved concentrations must be filtered prior to submission with a 0.45µm filter at the time of sample collection according to the procedure in 5.4.2.

Table 5.3.A. Sample container, hold time and preservative for common water chemistry parameters. Subject to change, verify with VAEL prior to sample collection.

Parameter	Sample Container	Hold Time (days)	Preservative
Chloride	50 mL plastic green cap	28	None
IC Anions - Sulfate	50 mL plastic green cap	28	Cool 4 deg C
Nitrogen – Ammonia	50 mL plastic blue cap	28	H ₂ SO ₄ to pH <2
Nitrogen – NOX	50 mL plastic blue cap	28	H ₂ SO ₄ to pH <2, Cool 4 deg C
Nitrogen Total	50 mL plastic blue cap	28	H ₂ SO ₄ to pH <2
Organic Carbon Dissolved	125 mL glass amber	28	Cool 4 deg C

Phosphorus (Total or Dissolved)	60 mL glass vial	22	None
Metals (Total or Dissolved)	125 mL round plastic	180	HNO ₃ to pH <2
Alkalinity	250 mL square plastic	14	Cool 4 deg C
Conductivity	125 mL round plastic	28	Cool 4 deg C
Gran Alkalinity/Conductivity	250 mL round plastic	10	Cool 4 deg C
Total Suspended Solids	1 L plastic	7	Cool 4 deg C
Turbidity	250 mL square plastic	2	Cool 4 deg C

Table 5.3.B Sample containers needed for analyzing pesticide concentrations in water samples. All samples analyzed for pesticides need to be kept cold at 0 – 6 degrees Celsius. Subject to change, verify with VAEL prior to sample collection.

Parameter	Sample Container
Neonicotinoids Corn Herbicides (including Atrazine)	1-L acetone-rinsed amber glass bottles with PTFE caps
Glyphosate	50-mL green-capped centrifuge tubes

5.3.2 Dissolved Concentrations - Field Sample Filtering

Filtering of an ambient water sample should be done at the time of sample collection prior to field preservation unless the program QAP indicates otherwise. Filtering is necessary to remove solids from the ambient water collected. Dissolved concentrations (dissolved phosphorus, dissolved nitrogen, dissolved metals, dissolved organic carbon, etc.) must be filtered prior to analysis.

Field Filtering Procedure:

1. Rinse filtering apparatus, filter, syringe, etc. with distilled water 3 times.
2. Rinse syringe with ambient water 3 times.
3. Place clean filter paper (0.45 µm mesh) using clean forceps inside filtering apparatus. Close filter snugly.
4. Fill syringe with ambient sample water and purge filter with ~10 mls of water without collecting the filtered water.
5. Place filtering apparatus above container and purge 10 mls into container. Use this water to “rinse” container with filtered water two times prior to filling. Do not rinse container with filtered water for dissolved phosphorus analysis.
6. Fill sample bottle with filtered water to provide the required sample volume to VAEL.

5.3.3 Dissolved Oxygen – Full Bottle Technique

1. Collect the dissolved oxygen sample using either a grab or a Kemmerer. The hose from the Kemmerer should be inserted to the bottom of the dissolved oxygen (D.O.) bottle and the glass sample bottle should be allowed to overflow twice the volume and then capped with the glass stopper. Care must be taken to prevent turbulence and the formation of air bubbles when collecting the sample and while filling the bottle.
2. Record the temperature of the water sample; the solubility of dissolved oxygen is temperature dependent.
3. Remove the glass stopper from the D.O. bottle and immediately add 2 ml of manganous sulfate solution (labeled “#1”), followed by 2 ml of alkaline iodide-azide solution (labeled

“#2”), below the surface of the liquid. Re-stopper the bottle with care to exclude air bubbles and mix well by inverting the bottle 8 to 10 times.

4. When the precipitate has settled leaving a clear liquid on top, shake again by inverting the bottle 8 to 10 times.
5. Store the sample in the dark for up to 8 hours prior to analysis.

5.3.4 Bacteriological Samples

The VAEL biology section accepts bacteria samples on specific days. Notify the laboratory at least one day in advanced of sample arrival to allow proper preparation of the media.

Procedure for sample collection:

1. Collect samples in sterilized 120 ml or 250 ml plastic bacteriological bottles. Do not remove the caps until sampling begins. Do not allow contamination of the inside of the bottle or cap.
2. Use special sample bottles containing sodium thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3$) for chlorinated samples. These bottles are sealed 250 ml IDEXX bottles.
3. Do not rinse bottles prior to sample-collection.
4. Remove cap from pre-labelled sterile bacteria bottle and grasp at base with one hand. Do not rinse bottle or touch rim of the bottle.
5. If sampling in flowing waters, locate the centroid of flow as the sampling location and allow any disturbance to flow downstream prior to sampling. Take samples mid-way between the surface and bottom of the stream. Facing upstream, plunge bottle mouth down into the water to avoid introducing surface scum. If possible, sample between 15 to 30 cm (6 to 13 inches) below the water surface, or at the depth relevant to the water under evaluation.
6. If the water body is static, an artificial current can be created by moving the bottle horizontally in the direction it is pointed and away from the sampler. Position the mouth of the bottle into the current away from the hand of the collector and away from the side of the sampling platform or boat.
7. Tip the bottle slightly upwards to allow air to exit and the bottle to fill.
8. After removal of the bottle from the water, adjust the water level to match the fill line indicated on the shoulder of the E. coli bottle.
9. Securely replace cap on bottle then ice samples until analysis.

Procedure for sample submission:

1. The laboratory needs prior notice before submitting bacteria samples. Samples should arrive at the Laboratory within 6 hours of sample collection.
2. Separate containers are required for each test requested. An inch of headspace is required to insure proper mixing.
3. Samples must be kept on ice from the time of sampling until refrigerated at the laboratory.
4. Samplers are instructed to use sterilized 120 ml sterile sealed containers unless dechlorination is necessary.
5. Chlorinated samples must be dechlorinated at the time of collection. On request, the laboratory will provide sterile 250 ml containers containing sodium thiosulfate. The sodium thiosulfate in the sample bottle will neutralize 250 ml of water containing up to 15 ppm chlorine.

5.3.5 Chlorophyll-A

Collect chlorophyll-a samples using an appropriate sampling method and keep samples on ice until filtered. Appropriate sampling methods are described in Section 5.1 Manual Water Sample Collection Procedures. Refer to the [Lay Monitoring Program Manual](#) (VTDEC 2013) for the filtering procedure using the vacuum pump and filtering apparatus.

5.3.6 Mercury

This method describes the collection technique for trace-metal clean sampling for mercury or other sensitive parameters. Samples should be collected upstream or beyond contact with the boat to avoid contamination by metal equipment.

Equipment:

- Tyvek suits
- Teflon Kemmerer
- Cooler
- Shoulder-length disposable gloves
- Waterproof sample labels
- Latex gloves
- Trace grade HNO₃
- Pipet (5 ml capacity)
- Disposable pipet tips
- Teflon sample bottles (double bagged in zippered plastic bags)

5.3.6.2 Surface Grab Procedure

1. Collection of aqueous mercury samples will conform to the EPA Method 1669 'clean hands-dirty hands' techniques (USEPA, 1996).
2. In brief, sampling staff will wear clean Tyvek windsuits and gloves. 'Clean hands' will be assisted by 'dirty hands' in all aspects of sampling to ensure that clean hands does not touch anything but the Teflon Kemmerer while sampling. It is especially important to avoid all metal, such as the side of an aluminum boat, pens, jewelry, clipboard, etc. Mercury has an affinity for metals, especially gold, and will potentially contaminate the sample if touched.
3. 'Clean hands' will wear shoulder-length gloves. Gloves will be new from the box at the time they are put on. Aqueous mercury samples will be stored in a separate cooler from other samples collected.
4. Using a clean 500 ml Teflon bottle, collect a water sample using a surface grab. Do not rinse.
5. Preserve samples in situ with 3.6 mls concentrated trace-metals grade HNO₃, using a new pipet tip rinsed twice in mercury-clean 10% HCl, and once in trace metal grade HNO₃.

5.3.6.2 Epilimnetic Grab Sampling

1. Waterproof sample labels are prepared using waterproof ink.
2. 'Dirty hands' opens the 'clean box,' gloves, and dons a Tyvek suit.
3. 'Dirty hands' removes shoulder glove and assists 'Clean hands' in donning shoulder-gloves and shorter gloves if necessary. From this point forward, 'Clean hands' handles nothing but the sample bottle, or the inner Ziploc bag which contains the sample bottle.

4. 'Dirty hands' opens the 'clean cooler,' and removes one 1000ml double bagged bottle. 'Dirty hands' opens the outer bag. 'Clean hands' reaches into the outer bag, opens the inner bag, removes the bottle, and folds the inner bag over. 'Dirty hands' seals the outer bag and replaces it into the 'clean cooler.'
5. 'Dirty hands' removes the autopipette from the clean cooler and affixes a new pipet tip. 'Dirty hands' rinses the pipet tip two times in reagent-water dilute 10% HCl, and one time in HNO₃. Rinsates are evacuated into a waste-acid container.
6. 'Clean hands' opens the sample bottle, evacuates the contents, and closes the bottle.
7. 'Clean hands' submerses the bottle to a 0.5-meter minimum depth, opens the bottle, and fills it 1/3rd full. The bottle is then surfaced, shaken, opened, and the rinsate evacuated away from the immediate sampling point. The bottle is resealed. This is repeated twice.
8. 'Clean hands' re-submerses the bottle and allows the bottle to fill entirely. The bottle is recapped underwater. 'Clean hands' surfaces the bottle and opens the cap slightly.
9. 'Dirty hands' draws 3.6 mls trace-metal grade HNO₃, and pipets this into the sample bottle. 'Clean hands' then tightly caps the bottle. 'Dirty hands' opens the clean cooler, withdraws, then opens the outer bag.
10. 'Clean hands' unfolds the inner bag, replaces the bottle, and seals the inner bag. 'Dirty hands' then seals the outer bag, affixes the label, and replaces the double-bagged sample in the clean cooler.

5.3.6.3 Hypolimnetic Kemmerer Sampling

1. Use an all-Teflon Kemmerer sampler to collect a sample one meter above the sediment water interface. The sampler should be rinsed three times in the lake water prior to collecting the sample.
2. 'Dirty hands' un-bags the double-bagged Teflon Kemmerer, affixes the line, and rinses the sampler three times in lake water by submersing the sampler, forcefully retrieving it, and allowing it to drip off. The sampler is then lowered 2 meters below the boat and tied off.
3. 'Dirty hands' opens the 'clean cooler,' and removes a 500ml double bagged bottle. 'Dirty hands' opens the outer bag.
4. 'Clean hands' reaches into the outer bag, opens the inner bag, removes the bottle, and folds the inner bag over. 'Dirty hands' seals the outer bag, and replaces it into the 'clean cooler.'
5. 'Dirty hands' lowers the Kemmerer sampler to 1 meter from the sediment-water interface and trips the closure mechanism with the non-metallic messenger. The sampler is retrieved.
6. 'Clean hands' opens and evacuates the bottle. 'Dirty hands' directs the sample stream from the Kemmerer sampler to fill the bottle 1/3rd full. 'Clean hands' caps the bottle, shakes vigorously, and evacuates the rinsate. This is repeated two times.
7. 'Dirty hands' directs the sample stream to fill the bottle entirely. 'Clean hands' caps the bottle, and 'Dirty hands' re-submerses and ties the Kemmerer sampler to hang in the water at 2 meters of depth.
8. 'Dirty hands' draws 1.8 mls trace-metal grade HNO₃ and pipets this into the sample bottle which was opened by 'Clean hands'. 'Clean hands' tightly caps the bottle.
9. 'Dirty hands' opens the clean cooler, withdraws the bag and then opens it.
10. 'Clean hands' unfolds the inner bag, replaces the bottle, and seals the inner bag. 'Dirty hands' then seals the outer bottle, affixes the label, and replaces the bottle in the clean cooler.

11. 'Dirty hands' bags the Kemmerer sampler with new bags, using "Clean hands" assistance.

5.3.6.4 Sediment Sampling for Mercury Analysis

Due to high mercury concentrations found in lake sediments, strict mercury-clean techniques will not be required, provided that cores are sectioned in the field, as soon as practical after collection. Project staff will, however, use gloves and an adapted clean hands-dirty hands protocol when collecting and sectioning sediments.

Equipment:

- Clean lot-certified PETE bottles, 125ml
- Glew-design modified KB corer with a 60 cm by 7 cm Lexan core tube, or a KB corer with a 60 cm by 5cm Lexan tube and a cellulose acetate butyrate liner.
- Core rubber caps
- Large plastic bags (to store Lexan tubes)
- Scraper
- Lexan sectioning tray
- Sonar
- Disposable powder-free gloves
- Zip-style plastic bags

Preparation and storage of field equipment: Acid clean the Lexan core tubes before going into the field. Core tubes will be stored in doubled, plastic bags between acquisitions. These bags will be replaced regularly. Core tubes will be acid washed every tenth sample collected, or when the field coordinator determines that cleaning is necessary. Core sectioning tools (scraper, Lexan sectioning tray) will be cleaned following the same schedule as core tubes and will be stored in plastic as well.

Sampling Procedure:

1. Once positioned at the lake sampling station, rinse tubes copiously in lake water prior to use.
2. Label two sample bags. The inner bag with a grease pencil, the outer bag with an adhesive label marked with indelible ink.
3. 'Clean hands' and 'dirty hands' are designated.
4. 'Dirty hands' assists "Clean hands" in donning gloves. 'Clean hands' rinses and handles the core tube, placing it into the corer head. 'Dirty hands' is responsible for handling the corer head and line, and for collecting the core. The core descent is tracked using SONAR.
5. When corer is just above lake sediments, release corer until the descent stops. Release messenger, wait for trigger to release and retrieve corer slowly.
6. 'Clean hands' caps the core bottom upon its arrival at the surface. Do not pull corer from water until bottom is capped. 'Dirty hands' removes corer head, while 'clean hands' holds the core. 'Clean hands' caps and sets the core to a rack, 'Dirty hands' assembles extrusion equipment.
7. 'Clean hands' places the core onto the extruder. 'Clean hands' affixes sectioning tray onto the core tube. 'Dirty hands' tightens associated fasteners. 'Clean hands' prepares sample bottles and removes sectioning tools from their bags. While 'dirty hands'

controls extrusion from the core bottom, 'clean hands' sections the sediment into the sample bottle. Bottles are placed singly into zip-style bags.

8. Sectioning sediment samples for dated sediment cores.
 - a. Rinse a Lexan sectioning tray copiously.
 - b. For those depths from which mercury samples are to be analyzed, the extruded sediments will be split on the tray, and each half moved into a new, clean Whirl-Pak-type bag using a plastic scraper.
 - c. Sediments will be stored in double bags, in a specially designated cooler. At no time will sediment samples be placed into the same cooler as aqueous mercury samples.
9. Rinse all field equipment (corer, tubes, scraper, and Lexan sectioning tray) in lake water after sediments are removed.
10. Sample Acceptance/Rejection: Cores will be rejected and the core re-collected if: 1) sediments contact metal portions of the corer head (overflow); 2) the sediment-water interface is disturbed; 3) the field coordinator judges that a contamination may have occurred, or the core is of poor quality; or 4) gaseous ebullition caused by temperature differential causes the core to break apart before sectioning.
11. Two cores will be acquired from each sampling station. The core reflecting the least disturbance will be selected for analysis at the laboratory.

5.4 Submitting Samples for Analysis

5.4.1 General Procedure

After collection and if needed, preservation, chemical samples are submitted to VAEL. All samples must be properly labeled and submitted as soon as possible after collection to allow for analysis to occur within the holding time. The VAEL Field Sheet is to accompany samples collected which contains the program information, date and time of collection, name of collector, list of samples collected, and any relevant information regarding temperature and preservation. Samples are to be submitted to VAEL Sample Receiving during normal business hours or submitted to the first floor holding refrigerator if after business hours. If submitting samples in the first floor holding refrigerator, complete the sample submission form including the temperature of the samples and time submitted. Notify sample receiving staff via email at agr.vael@vermont.gov.

5.4.2 Chain of Custody

Chain of custody procedures are necessary to ensure the legal integrity of sample materials collected and submitted to the VAEL for analysis. Chain of custody procedures should be followed for samples that will be subject to higher-than-normal scrutiny, such as samples related to enforcement cases. The validity of the test results is assured if the Department can show that the samples collected were maintained safe from tampering or interaction with contaminating chemicals. This requires the complete written documentation of the security of the sample to be recorded from collection to disposal. For samples requiring chain of custody documentation and submission, refer to the [VAEL QSM](#) and the Watershed Management Division's Standard Operating Procedure (SOP) for Determining Water Quality Impacts to Rivers and Streams for Environmental Enforcement Cases (in draft).

6 Biological Sampling Methods

6.1 Phytoplankton

6.1.1 Vertical Qualitative Sampling-Plankton Net

Equipment:

- Wisconsin plankton net (63 mm mesh) with calibrated line
- Clear glass 50 mL tube
- Lugol's iodine preservative
- Secchi disk
- Depth tape
- Squirt bottle
- Cooler

Procedure:

1. Determine the depth at the sampling site. Vertical qualitative samples can be obtained in waters greater than 3 m in depth.
2. Determine the Secchi disk transparency.
3. Rinse out the plankton net with ambient surface water and close stopcock.
4. Lower the net to the desired depth (i.e. twice the Secchi and at least one meter from the bottom).
5. Retrieve slowly ($\frac{1}{2}$ meter per second) and allow excess water to drain through the mesh sides. Release the sample through the stopcock opening into the amber bottle or centrifuge tube.
6. Rinse the net 3 times with stopcock open without going over the top, close stopcock, drop to desired depth and retrieve. Rinse the outside of the net with ambient water to get organisms stuck to the net in the bucket. Do not rinse the inside of the net. Concentrate the sample to fit into a 50ml container and preserve.
7. Repeat the vertical hauls until an adequate amount has been collected. This will vary with project needs.
8. Record information on bottle label: date, time, location, depth of haul, type of sample (e.g., vertical-qualitative).
9. Add Lugol's preservative (2 ml/L or 5 drops per 50 ml tube), secure container tightly and invert to mix thoroughly.
10. Store sample in a cool, dark place, such as a cooler.

6.1.2 Horizontal Qualitative Sampling

Equipment:

- Wisconsin plankton net (63 mm mesh) with calibrated line
- Clear glass 50 mL tube
- Lugol's iodine preservative
- Secchi disk
- Depth tape
- Collection bucket (>4 liter)

- Squirt bottle
- Cooler

Procedure:

1. Rinse out the plankton net with ambient surface water and close stopcock.
2. Rinse collection bucket with ambient surface water.
3. Throw the net from a stationary point and slowly retrieve until an adequate sample has been collected or tow the net slowly behind a boat until an adequate sample has been collected.
4. Prevent the net from touching the bottom, particularly when sampling from shore. Note depth in meters on rope.
5. Rinse the net 3 times with stopcock open without going over the top, close stopcock, drop to desired depth and retrieve. Rinse the outside of the net with ambient water to get organisms stuck to the net in the bucket. Do not rinse the inside of the net. Concentrate the sample to fit into a 50ml container and preserve.
6. Record information on bottle label: date, time, location, type of sample (e.g. horizontal-qualitative), and length of haul.
7. Add Lugol's preservative (2 ml/L or 5 drops per 50 ml tube,) and invert container to mix thoroughly.
8. Store sample in a cool, dark place, such as a cooler.

6.1.3 Composite Quantitative Sampling-Hose

Equipment:

- Sampling hose
- Clear glass 50 mL tube
- Lugol's iodine preservative
- Secchi disk
- Depth tape
- Cooler
- Collection bucket (>4 liters)

Procedure:

1. Determine the Secchi disk transparency and the depth at the sampling site.
2. Rinse collection bucket with ambient water.
3. Collect a hose sample at twice the Secchi transparency depth or to within one meter of the bottom, whichever is less.
4. Empty the entire contents of the hose into the rinsed collection bucket and swirl to mix.
5. Pour a 50 mL subsample into the glass tube.
6. Record information on bottle label: date, time, location, depth of composite, and type of sample (e.g., composite - quantitative).
7. Add Lugol's preservative (2 ml/L or 5 drops per 50 ml tube), and invert container to mix thoroughly.
8. Store sample in a cool, dark place, such as a cooler.

6.1.4 Discrete Quantitative Sampling-Kemmerer

Equipment:

- Kemmerer sampler, if necessary
- Clear glass 50 mL tube
- Lugol's iodine preservative
- Cooler

Procedure:

1. Collect sample from a discrete depth using a Kemmerer or from the surface by taking a grab sample.
2. Pour sample into glass tube. Record information on label: date, time, location, and type of sample (e.g., discrete sample - quantitative), depth of haul.
3. Add Lugol's preservative (2 ml/L or 5 drops per 50 ml tube), and invert container to mix thoroughly.
4. Store sample in a cool, dark place, such as a cooler.

6.1.5 Discrete Quantitative Sampling- Plankton Net

Note: This method is used primarily by the Lake Champlain Monitoring Program to collect blue-green algae.

Equipment:

- Wisconsin plankton net (63 μ m mesh) with calibrated line
- Clear glass 50 mL tube
- Ethyl alcohol (ethanol or ETOH) (preservative)
- Secchi disk
- Collection bucket with stopcock (e.g. 2 gallon water cooler)
- Cooler

Procedure:

1. Rinse the collection bucket in ambient water.
2. Rinse the plankton net with surface water and close stopcock.
3. Throw the net from a stationary point and slowly retrieve or tow the net slowly behind a boat until an adequate sample has been collected. Prevent the net from touching the bottom, particularly when sampling from shore.
4. Rinse the net 3 times with stopcock open without going over the top, close stopcock, drop to desired depth and retrieve. Rinse the outside of the net with ambient water to get organisms stuck to the net in the bucket. Do not rinse the inside of the net. Concentrate the sample to fit into a 50ml container and preserve.
5. Record information on bottle label: date, time, location, type of sample (e.g. horizontal - qualitative) and length of haul.
6. Add Lugol's preservative (2 ml/L or 5 drops per 50 ml tube) and invert container to mix thoroughly.

7. Store sample in a cool, dark place, such as a cooler.

6.1.6 Sediment Diatoms

Vermont adopted the methods used in the 2007 and 2012 National Lake Assessment to collect sediment diatoms. The latest description of the sampling methods used is from the 2017 National Lake Assessment Northeast Regional Voucher Flora Sampling Protocols and is copied below.

Equipment:

- KB sediment corer with messenger
- Core tubes with plugs
- Quart-sized plastic sealable bags (3 per lake)
- Whirl-Pak bags
- GPS unit
- Anchors
- Depth sounder
- Surgical gloves
- Sharpie
- Labels
- Packing tape
- Squirt bottle with DI water
- Syringe
- Plastic spatula
- Cooler with ice
- Large clean plastic garbage bags

Procedure:

Collect a sediment core of at least 5cm from undisturbed sediments at or near the index site and section off both the 0-1 cm and 1-2 cm of sediment from the top of the core sample for analysis. Sediment from the 0-1 cm and 1-2 cm cookies will be kept in separate labeled quart-sized plastic sealable bags.

In natural lakes, the composition and texture of the bottom will vary from lake to lake and, in some lakes, it will be impossible to get a 5 cm core because the bottom is too rocky, the sediments are too dense, or, if it is a shallow lake, there are macrophytes covering the bottom. It is essential that the GPS coordinates be recorded and the collection location be marked on the northeast regional voucher flora sediment diatom sample collection form.

If you collect a core less than 5 cm long on your first try, move to another location near the index site to find an area with a softer bottom. In addition, you can experiment with getting improved penetration by adding additional weight (if available) to the corer, and/ or by releasing the corer further above the sediments. If a 5 cm core sample cannot be collected from natural lakes after attempting at least three locations, process the last core that you obtained. The procedures for collecting and processing sediment cores are presented below.

If you collect a core longer than 5 cm long, this will be acceptable if is water on top of the sediment core.

Preparing to take the core

1. To minimize the amount of swinging around the boat will do while sending the corer down and pulling it back up, it is advised that two anchors are used during sampling. Deploy the bow anchor into the wind, back up the boat until all or almost all anchor line is out. With the bow still pointed into the wind, throw or lower a second anchor off the stern of the boat. Move the boat forward while letting line out on the stern anchor and pulling line in on the bow anchor. When positioned roughly equally between the two anchors secure the bow anchor line to the bow and the stern anchor line to the stern.

2. In order to minimize the disturbance of the sediment, always drop the corer at least 5 m from the location you dropped the core in prior sediment sampling attempts. To do this you will likely need to let out line on one anchor while taking in line on the other anchor. If the boat is big enough, moving to another part of the boat can achieve this effect as well.

3. Taking a sediment core from a motorboat tends to be easier than from a canoe. However, keep in mind the height of your freeboard, you'll want to sample from the area of your boat with the least amount of freeboard in order that the person plugging the stopper in to the bottom end of the successfully collected core can reach into the water to put the plug in before the core breaches the surface.

4. Cores can be taken both from two canoes rafted together or from a single canoe. Rafting canoes together makes a more stable platform and is accomplished with two bars/rods/wooden posts clamped across the two canoes with a space of about 0.75 m between the two canoes. The core is then dropped between the two canoes with the person dropping the core standing and the person plugging the core leaning into the water while the crew members in the other canoe lean their weight away from the adjacent canoe to help provide more stability for the coring team. If only one canoe is taking a sample, it is best for both crew members to work from as far forward in the bow or as far back in the stern as possible. The crew member at the very end of the canoe is the one who leans into the water to plug the core while the other crew member stands while dropping and retrieving the core, being sure to shift their balance backwards as their crew mate leans into the water when the top of the core is breaching the surface.

5. Maine will be piloting coring from inflatable rafts and will develop additional guidance to others who may need to do this in future years. 6. If weather permits, the crew member in charge of plugging the core may find it is more pleasant and easier to do so from in the water while swimming.

Sediment Core Sample Collection

1. Wear surgical gloves at all times during sample collection to protect yourself from any potential contaminants in the sediments.



2. If the bottom has been disturbed during the initial depth determination or for any other reason, move at least 5 m to take the core. It is critical that the corer strikes undisturbed surface sediments.
3. Using the modified KB corer and core tube supplied by the NLA, insert the core tube into the sampling housing apparatus and tighten the 'yes' hose clamp screws to secure the tube. Ensure the messenger is attached to the sampler line. Set the release mechanism (i.e. pink lacrosse ball) (Figure 1).
4. Recheck your depth using an electronic depth sounder, do not use a tape and weight to check the depth as it may disturb the surface sediments we are about to collect.
5. Slowly lower the corer through the water column, keeping count of the depth lines on your line as you drop it. When you determine to the best of your abilities that the bottom of the core tube is 1 m above the sediment surface. Stop lowering the core and brace yourself and pick a time when the boat is most steady to drop the core straight up and down into the bottom substrate. Immediately after the corer drops into the sediments, maintain line tension to prevent the corer from tilting and disturbing the core sample. Keep in mind that the goal is to obtain the surface sediments as undisturbed as possible. If the 5 cm core length is not obtained the first time, the operation might need to be repeated at a new location using additional weights on the corer (if available) and/or a greater release height to improve penetration and obtain a longer or more level core. If the core length exceeds the length of the core tube and there is no water at the top of the core tube, the operation will need to be repeated at a new location using less weight on the corer and/or a shorter release height.
6. Trip the corer by releasing the messenger weight so that it slides down the line. Keeping the line vertical and keeping tension on the line will help ensure that the messenger reaches the sampler and trips the mechanism. Often you can hear and feel the messenger reaching its target and triggering the top plug to engage. Continue to keep tension on the line so as not to let the core tip to one side while the messenger is on its way down. Having two anchors deployed is advised in order to minimize the swinging around of the boat at this time.
7. Pull the corer up at a slow and steady pace, jerking and stopping can result in loss of material or mixing of the surface sediments. When the top of the corer becomes visible the crew member with the plug (the Plugger) will reach into the water as far as possible with their hand that has the plug and use their other hand to follow the core tube down as their crew mate (the Wincher) brings the core to the surface. While the Wincher keeps the bottom of the core tube under water, the Plugger will reach under the surface and plug the bottom of the corer with a corer tube plug. To do this without disturbing the water-sediment interface, neither crew member should tilt the corer more than 45 degrees. (Note: core tube plugs are easily lost. Be sure to have spares available at all times.)
8. Keeping your hand under the corer tube plug, raise the corer into the boat in a vertical position. Stand the corer in a large tub or bucket to prevent contaminating the boat with sediment material.
9. Be sure to wash all mud off the boat before you leave a lake or travel to another lake so as not to give any aquatic invasive species hijackers a ride!

Sediment Core Processing

This procedure can be done in the boat as required by the 2017 NLA sediment sampling protocol, or if waves and weather conditions merit, the core can be sealed with all the overlying water with orange caps at both ends and kept secured in an upright position and carefully transported to a calmer part of the lake or the nearest landing where the core can be extruded on land. During transport, it is very important to keep the surface of the core from mixing with the overlying water and deeper sediments. *(NOTE: This deviates from the NLA 2017 sediment contaminant protocol which prefers that you move to a more protected coring location and perform extrusions in the boat rather than taking the core to shore where there is risk the top 5cm may get mixed with deeper sediments.)*

1. Put gloves on. Record the Site ID, date, and collection intervals on sediment core sample labels. There are two sediment core intervals, 0-1 cm and 1-2 cm. Attach the labels to two plastic quart plastic sealable bags. Cover labels with clear packing tape.
2. Rinse the “sampling kit” with DI or tap water.
3. Detach the core tube from the corer. One crew member should hold the sampler in a vertical position while the second person dismantles the unit.
4. Position the extruder under the corer tube plug at the base of the coring tube. Supporting both the core tube and the extruder in a vertical position, slowly lower the coring tube onto the extruder until the sediment is approximately 1 cm below the top of the tube. As you lower the coring tube, the water will spill slowly over the edges of the top of the core (Figure 1).
5. Remove the remaining water above the sediment core by using a syringe with tube so that the surface sediments are not disturbed. Conversely, one can use a squirt bottle or turkey baster to carefully suction out the water without disturbing the sediments.
6. Secure the sectioning stage onto the top of the coring tube. Place the Plexiglas sectioning tube on the stage directly over the coring tube (Figure 2). Slowly extrude the sediment core into the sectioning tube until the top of the sediment reaches the 1 cm line on the sectioning tube (Figure 3). Slide the sectioning tube onto the flat part of the stage and scrape the top 1 cm section of sediment with the spatula into the plastic sealable bag or Whirl-Pak labeled for the 0-1 cm. This sample will often be very soupy. Seal the sample bag and put in cooler with ice or ice packs.
7. Remove the stage and sectioning tube and rinse in lake water. Also rinse the spatula, gloved hands and any other implements that have come in contact with the sediment. This procedure prevents contamination of the 0-1 cm layer with diatoms from the 1-2 cm portion of the core. Rinse any sediment from your gloved hands.
8. Re-secure the sectioning stage onto the top of the coring tube. Place the Plexiglas sectioning tube on the stage directly over the coring tube (Figure 2). Again, slowly extrude the sediment core into the sectioning tube until the top of the sediment reaches the 1 cm line on the sectioning tube (Figure 3). Slide the sectioning tube onto the flat part of the stage and scrape the top 1 cm section of sediment with the spatula into the plastic sealable bag labeled for the 1-2 cm sample. Seal the sample bag and put in cooler with ice or ice packs.

9. Discard the rest of the core (unless you are planning to take bottom cookies or preserving the entire core). Rinse the corer, spatula, coring device, and sectioning apparatus thoroughly with lake water. Rinse with tap water and phosphate free detergent (e.g. Alconox) at the next base site. After cleaning the core tube, cover the ends with the orange caps and place all other sectioning equipment into a clean plastic bag.
10. Keep samples cool until you can transfer them to a freezer kept at -10 to -20 degrees C. Samples will stay in the freezer until all lakes are completed and they can be shipped for analysis.

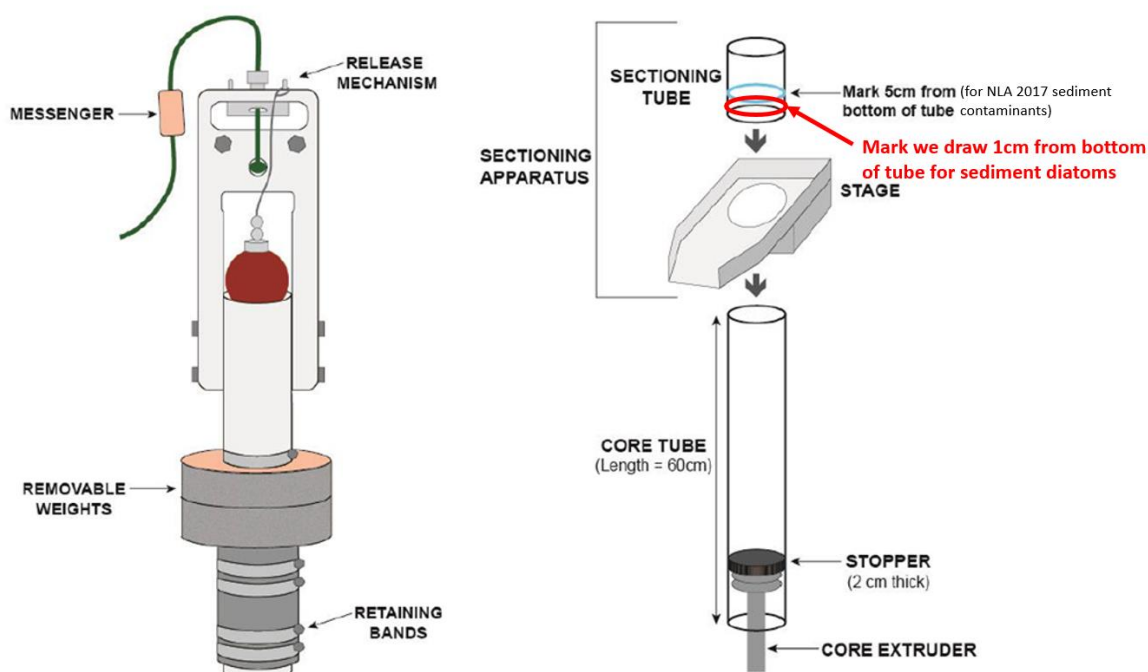


Figure 6.1.A Illustration of the core tube and sectioning apparatus. Note, field crews need to draw the mark denoting the 1 cm mark on the NLA 2017 sectioning tube.



Figure 6.1.B Setting sectioning tube with 1 cm marking over stage. Water has already been siphoned off the top of the core.



Figure 6.1.C 0-1 cm cookie extruded to 1 cm line

6.2 Zooplankton

6.2.1 Vertical Qualitative Sampling-Plankton Net

Equipment:

- Wisconsin plankton net (63 μ m mesh) with calibrated line
- Clear glass 50 mL tube
- CO₂ water (seltzer)
- Ethyl alcohol (ethanol or ETOH) or formalin sugar (preservative)
- Secchi disk
- Depth tape
- Collection bucket with stopcock (e.g. 2 gallon water cooler)
- Cooler

Procedure:

1. Determine the depth at the sampling site. Vertical qualitative samples can be obtained in waters greater than 3 meters in depth.
2. Determine the Secchi disk transparency.
3. Rinse the collection bucket in ambient surface water.
4. Rinse out the plankton net and close stopcock.
5. Lower the net to the desired depth (i.e. twice the Secchi and at least one meter above the bottom).
6. Retrieve slowly (0.5-1.0 m/sec) and shake out excess water. Drain the sample through the stopcock opening in the collection bucket into the amber bottle or centrifuge tube.
7. Repeat the vertical hauls until an adequate sample has been collected.
8. Record information on bottle label: date, time, location, depth of haul, type of sample (e.g., vertical qualitative) and mesh size.
9. Add a small amount of seltzer (carbonated water) to reduce distortion observed under a microscope.
10. Add enough preservative to create a 70% ethanol final concentration (50% of sample with 95% ethanol). Invert container to mix thoroughly.
11. Store sample in a cool, dark place, such as a cooler.

6.2.2 Vertical Quantitative Sampling for trace metal burdens in zooplankton

Equipment:

- Metal-free Wisconsin-style net with “cod” end or equivalent (with appropriate mesh size, see sections below)
- 200 ml lot-certified PETE ‘compositing vessel’
- 500 ml acid cleaned squirt bottle (Clean after every tenth sampling event).
- 500 ml squirt bottle for CO₂ water (seltzer)
- CO₂ water (seltzer)
- 1 pre-weighed, pre-coded, lot-certified 50 ml polycarbonate sample vessel
- 2 non-weighed 50 ml polycarbonate vessels

- powder-free vinyl gloves
- protective plastic sheet, 4'x 4' or larger
- field sheet
- 200 ml lot-certified PETE 'compositing vessel'
- formalin solution (preservative)
- Depth tape
- Dolphin bucket (201 micron mesh)
- Cooler

General Procedure:

1. Prior to going out into the field, a pre-coded 50 ml sample vessel is weighed to the nearest 0.001 g, and the weight and code recorded.
2. In the field, after the vessel has arrived at station and has been securely anchored, 'Clean hands' and 'Dirty hands' are designated. 'Clean hands' and 'dirty hands' don regular-length powder-free vinyl gloves.
3. A plastic sheet is draped over the gunwale of the sampling boat, such that the net will not have the opportunity to contact the boat.
4. 'Dirty hands' removes and assembles the non-metallic net, and 'clean hands' and 'dirty hands' jointly backflush the net three times in lake surface water. The dolphin bucket is similarly rinsed.

Procedure for Tows of Total Mercury (HgT) and Biomass Determination:

1. 'Dirty hands' lowers the net to within 1 meter of the lake bottom, and rests the net 30 seconds to allow the water column to recolonize.
2. 'Dirty hands' records the depth of this tow on the field sheet.
3. 'Dirty hands' retrieves the net at a rate of < 1 m/second.
4. When the net-hoop breaches the surface, 'dirty hands' lifts the net, and rinses the contents down along the net-sides using lake water and an acid cleaned squeeze bottle.
5. Once the sample is condensed into the dolphin bucket, 'clean hands' removes the bucket, further reduces the sample, and decants it into the 200 ml 'compositing vessel'.
6. This tow collection procedure is repeated until a minimum of 5 tows are collected. The field coordinator will determine if additional tows are necessary to obtain sufficient material for biomass and HgT analyses.
7. The contents of the compositing vessel are decanted to the dolphin bucket, and the contents reduced to < 50 ml volume.
8. 'Clean hands' opens the 50 ml sample vessel, rinses it three times with lake water, and decants the reduced composite plankton material into the vessel. The vessel is then filled to 50 ml with lake water, and capped tightly.
9. 'Dirty hands' opens a zippered bag, and 'clean hands' gently drops the filled 50 ml vessel into the bag.
10. 'Dirty hands' closes the bag and places it into the designated cooler for submission to the laboratory for analysis.

Procedure for tows for Taxonomic Analyses 201 micron mesh:

1. Two additional tows are composited, using the 201 m mesh, into the compositing vessel using the procedure outlined above.
2. The contents of the compositing vessel are then covered with seltzer water, capped, and allowed to sit 1 minute. At this time, the contents are returned to the dolphin bucket, reduced to the maximum extent possible, rinsed using the seltzer-squeeze bottle into a labeled 50 ml sample vessel, to approximately 25 ml volume.
3. The sample is capped and allowed to sit 5 minutes. The sample is then opened, and filled to 50 ml with formalin-solution. Invert container to mix thoroughly
4. Store sample in a cool, dark place, such as a cooler.

Tows for Taxonomic Analyses - 45-200 micron mesh:

1. Two tows are composited using the 45 micron mesh net, following the procedure outlined directly above.
2. While the dolphin bucket is held above the assembled 45 micron net, the contents of the 45 m composite are passed through the dolphin bucket, and allowed to run out into the 45 m net. This step removes plankton in the 201 micron fraction from the 45 -200 micron fraction.
3. The 45 micron sample is then recondensed, and transferred back to the compositing vessel.
4. The contents of the compositing vessel are then covered with seltzer water, capped, and allowed to sit 1 minute. At this time, the contents are returned to the dolphin bucket, reduced to the maximum extent possible, rinsed using the seltzer-squeeze bottle into a labeled 50 ml sample vessel, to approximately 25 ml volume.
5. The sample is capped and allowed to sit 5 minutes. The sample is then opened, and filled to 50 ml with formalin-solution.
6. Store sample in a cool, dark place, such as a cooler.

6.2.3 Horizontal Qualitative Sampling-Plankton Net

Equipment:

- Wisconsin plankton net (63 μ m mesh) with calibrated line
- 250 ml amber glass bottle with label (long-term storage) or plastic 50 ml centrifuge tube (short-term storage)
- CO₂ water (seltzer)
- Ethyl alcohol (ethanol or ETOH) or formalin sugar (preservative)
- Secchi disk
- Collection bucket with stopcock (e.g. 2 gallon water cooler)
- Cooler

Procedure:

1. Rinse the collection bucket in ambient water.
2. Rinse the plankton net with surface water and close stopcock.

3. Throw the net from a stationary point and slowly retrieve or tow the net slowly behind a boat until an adequate sample has been collected. Count the lines one meter apart on the rope. Prevent the net from touching the bottom, particularly when sampling from shore.
4. Shake out excess water and drain the sample through the stopcock opening in the collection bucket into the amber bottle.
5. Record information on bottle label: date, time, location, type of sample (e.g. horizontal - qualitative) and length of haul.
6. Add a small amount of seltzer (carbonated water) to reduce distortion observed under a microscope.
7. Add enough preservative to create a 70% ethanol final concentration. Invert container to mix thoroughly.
8. Store sample in a cool, dark place, such as a cooler.

6.2.4 Composite Quantitative Sampling-Plankton Net

Equipment:

- Wisconsin plankton net (63 μ m mesh) with calibrated line
- Clear glass 50 mL tube
- CO₂ water (seltzer)
- Ethyl alcohol (ethanol or ETOH) or formalin sugar (preservative)
- Depth tape
- Cooler

Procedure:

1. Determine the depth at the sampling site.
2. Rinse out the plankton net and close the stopcock.
3. Lower the net until it is one meter above the bottom or other appropriate depth.
4. Pull the net to the surface using a slow steady retrieve (0.5-1.0 m/sec). Keep the net vertical.
5. Shake out excess water and drain the sample into an amber glass bottle or centrifuge tube.
6. Rinse the net before attempting the next haul, collecting rinse water into the sample. This prevents loss of organisms back into the surface water as it descends the water column.
7. Collect a minimum of three hauls, all emptied into the sample amber bottle (composite) or in separate bottles for discrete replicates).
8. Record information on bottle label: date, time, location, depth of composite, number and length of hauls, diameter of net opening, and type of sample (i.e., composite-quantitative) and mesh size.
9. Add a small amount of seltzer (carbonated water) to reduce distortion observed under a microscope.
10. Add enough preservative to create a 70% ethanol final concentration. Invert container to mix thoroughly.
11. Store sample in a cool, dark place, such as a cooler.

6.2.5 Discrete Quantitative Sampling-Schindler or Juday Trap

Equipment:

- Schindler or Juday plankton trap (63 μ m mesh net) with calibrated line
- Amber glass bottles with labels or plastic 50 ml centrifuge tubes (short-term storage)
- CO₂ water (seltzer)
- Ethyl alcohol (ethanol or ETOH) or formalin sugar (preservative)
- Cooler

Procedure:

1. Rinse out the net of the sampling device with surface water and close the stopcock.
2. Lower the sampling device to the desired depth, close device if required, and retrieve the sample.
3. Shake the excess water from the collection net and drain the sample into an amber glass bottle.
4. Record information on bottle label: date, time, location, type of sampling device (e.g. 25 L Schindler), depth of sample and type of sample (e.g. discrete - quantitative). Add a small amount of seltzer (carbonated water) to reduce distortion observed under a microscope.
5. Add enough preservative to create a 70% ethanol final concentration. Invert container to mix thoroughly.

6.3 Macrophytes

6.3.1 Qualitative Aquatic Plant Surveys

Aquatic plant populations on selected lakes are identified, surveyed, mapped and summarized through qualitative (legacy) surveys. Purposes of the surveys are to: assess current plant densities, abundance and distribution; create a species list for each surveyed lake; document nuisance conditions or the degree of use impairment; monitor changes in plant populations over time; monitor plant response to control programs; evaluate effectiveness of control on selected species; and monitor populations of rare, threatened or endangered plants.

Plant Survey Types:

1. **Plant Survey:** A survey of the portion of the littoral zone visible from the water's surface, noting as many plant species as feasible (based primarily on visibility) and providing information on plant abundance, distribution, condition, depths (of certain species) and bottom type.
2. **Aquatic Invasive Species Search:** Lakes thought to be especially susceptible to watermilfoil, water chestnut or other non-native species introductions are checked on a rotating basis to identify new introductions as soon as possible. Field notes contain general descriptions of the amount of plant growth and nuisance conditions, but this type of survey does not provide a thorough list of all species present or their abundance.
3. **Aquatic Invasive Species Survey:** A preliminary survey or post management survey primarily of the non-native species in question, noting abundance, location, and depths of growth. Although areas of native species' growth are noted where they are mixed with the non-native species, this type of survey does not provide a thorough list of all species present or their abundance. Depending on the species and abundance found, a more formal Quantitative Aquatic Plant Point Intercept (PI) Surveys will be conducted by staff, or an accredited contractor.

Equipment:

- Boat and associated equipment
- Light anchor
- Aquatic plant sampling rake
- Depth tape
- Water scope
- Bucket
- Sealable plastic bags
- Cooler
- Detailed lake map
- Paper, pencil and clipboard
- Aquatic plant key



Procedure:

1. Slowly circle the entire perimeter of the lake, observing all areas less than 20 feet in depth.
2. Record the plant species seen and describe their relative abundance or density. According to the categories listed below. Note the depth of water that the plants are growing in. Sketch their location on a lake map. Each time a plant community changes, a new area should be delineated on the map. Be sure to map areas devoid of plants as well as areas with plants.

<u>Density Category</u>	<u>Cover</u>
Very abundant	75 - 100%
Abundant	50 - 75%
Common	25 - 50%
Occasional	5 - 25%
Scattered	1 - 5%
Uncommon	1%
Solitary	single plant

3. If waves or glare limit visibility, view plants through a water-scope or drag up plants with an anchor or rake to examine more closely.
4. When field identification is not possible, plants are placed in plastic bags, labeled, and transported to the Lakes and Pond Management and Protection Section for future identification. If kept overnight, the specimens are stored in a cool location to avoid deterioration. Dissecting microscopes, taxonomic keys and a reference collection are available at Lakes and Ponds to assist with identification.
5. Describe of each area in the completed report, including a species list for the lake and a brief general statement on the plant growth in the lake. This statement will provide a general observation of what was found, referring to dominant plants, areas of growth, possible problem areas and the necessity for plant control procedures. The more descriptive the statement, the more useful it will be in the future.

6.3.2 Quantitative Aquatic Plant Point Intercept Surveys

In 2014 the Lake Assessment Program adopted the quantitative aquatic plant point intercept (PI) method used by the Wisconsin Department of Natural Resources. The program follows the protocols outlined in [Recommended Baseline Monitoring of Aquatic Plants in Wisconsin](#) (Wisconsin Department of Natural Resources, 2010).

6.3.3 Littoral Habitat Assessment Macrophyte Sampling

While macrophytes are biological communities they are also habitat for other species. Refer to Section 3.8.2 to determine how macrophyte communities are sampled as part of Littoral Habitat Assessment.

6.3.4 BioBase Aquatic Plant, Bathymetric and Bottom Hardness Surveys

The BioBase EcoSound system is a cloud-based platform that processes raw sonar log files recorded using a Lowrance Sonar/GPS capable Chartplotter. Sonar data collected on the water is uploaded to an online account where it can be processed automatically using algorithms on BioBase's servers. The data is then used to create bathymetric, biovolume, and bottom hardness maps. These maps provide DEC and the public the necessary mapping to make informed decisions on lake management, including permitting, restoration, and recreation.

Bathymetric, biovolume, and bottom hardness surveys should occur in accordance with survey techniques used in the BioBase EcoSound System User Manual (Navico Holding AS 2020).

6.4 Periphyton

6.4.1 Periphytic Chlorophyll Accumulation

Periphytic chlorophyll accumulation is used as a simple indicator of stream primary productivity. This method involves the deployment of unpolished rectangular granite blocks attached to natural cobble substrate. The tiles are placed in the stream for a pre-determined period of time. Upon retrieval the periphyton accumulated on the tiles is removed, filtered and analyzed for chlorophyll-A.

This methodology is appropriate for medium to high gradient, hard-bottomed stream reaches, and is not applicable to large rivers or low gradient streams, or other aquatic ecosystems (e.g., lakes, wetlands). This method was specifically developed by the VTDEC for use in paired site comparisons for determinations of compliance with the [Aquatic Biota Criteria of the 2019 Indirect Discharge Rules](#), as revised (VTDEC 2019). Deployment of blocks for quantifying periphytic chlorophyll accumulation will typically be 14-21 days. The deployment will typically overlap with the second half of the six-week macroinvertebrate survey (artificial substrate deployment) outlined in Section 6.5.2.1, generally during late summer/early fall (or as otherwise specified in the indirect discharge permit). While this method has historically been used to bracket impact areas and spatial controls at indirect discharge sites, there may be additional utility for other types of studies.

Equipment:

- Rectangular granite blocks (0.25" x 1" x 3")
– minimum 14 per site
- Rubber bands (new, size 33 – 3.5" x 0.125")
- Natural cobbles, similarly sized – minimum 7 per site
- 125 ml plastic bottle – 12 per site
- Meter stick or depth tape
- Measuring tape
- Periphytic artificial substrate information field sheet



Procedure:

1. Attach two rectangular granite blocks to each of seven cobbles using two rubber bands (see photo). In streams being sampled for the first time, or streams prone to scour or sediment deposition, additional blocks and cobbles may be required to ensure that the minimum of 12 can be collected. Position blocks parallel to each other. Choose cobbles on site that have a relatively flat surface on at least one side for granite block attachment. Choose cobbles that are similarly sized, with the attachment surface similar or slightly larger than the combined surface area of the granite blocks.
 - a. Number blocks (1-14) and mark with the station name on the underneath surface with indelible ink, with consecutive number pairs (e.g., 1-2, 3-4) attached to each cobble.

2. Install the cobbles on the streambed with the granite blocks positioned on top.
 - a. Choose installation locations based on similarity of microhabitat, including flow velocity, depth, and canopy cover. As these methods are typically used for paired site comparisons (e.g., indirect discharge studies), similarity of microhabitat must also be considered and maximized between the paired stream sites.
 - b. Place cobbles in riffles comprised of similar coarse substrates. Installation locations should be in or near the thalweg, or other areas with sustained moderate flow. Placing cobbles directly adjacent to boulders or bedrock should be avoided.
 - c. Place cobbles directly on the surface of the streambed. This may require adjusting or moving other substrate to prepare the installation location for the cobble with the attached granite blocks. The cobble with the attached granite blocks can be worked into the streambed to a shallow depth to decrease the likelihood of being mobilized during a freshet flow event.
 - d. Place cobbles with numbered blocks consecutively and longitudinally in the channel. Cobbles are typically spaced several feet apart and can be placed adjacent to the artificial substrate baskets discussed in Section 6.5.2.1, if applicable to the study.
3. Record relative position in the channel, estimated canopy cover, measured depth of the granite blocks and estimated velocity for each installed cobble. Relative position in the channel can include a sketch of cobble locations and/or measurements taken from a fixed position adjacent to the channel to aid in locating cobbles for retrieval.
4. Cobbles with attached blocks are left in the channel for a period of 14-21 days and then retrieved. Upon retrieval of each cobble:
 - a. Record relative position, estimated canopy cover, measured depth and estimated velocity.
 - b. Assess the overall condition of the cobble and blocks using the following rating scale:
 - i. 1 – Undisturbed
 - ii. 2 – Some sediment and/or minimal movement. Depth, velocity and orientation maintained.
 - iii. 3 – Some sediment and/or significant movement. Depth, velocity and orientation altered.
 - iv. 4 – Significantly dislodged, was not submerged, and/or had poor orientation or was buried.
 - c. 12 blocks are collected for chlorophyll analysis. This will be the first 12 numbered blocks, unless cobbles with attached blocks have been lost entirely, or unless blocks with ratings of '3' can be replaced with more favorable numbers. Granite blocks with a rating of '4' should be rejected from the study.
5. Granite blocks are placed separately in 125 ml sample bottles containing stream water. Immediately place sample bottles on ice in a dark environment (cooler) until they are delivered to a lab for processing.
6. For paired site comparisons, granite blocks must be deployed and collected on the same day.
7. In the lab, periphyton is scraped from individual blocks, and the stream water and removed periphyton are filtered. Filters are frozen and analyzed for chlorophyll using Standard Methods 18 10200-H.

6.5 Macroinvertebrates

6.5.1 Lotic Semi-Quantitative Benthic Surveys

The riffle kick-net and multi-habitat sweep-net methods described here have been used in Vermont to collect consistent and replicable macroinvertebrate data since the late 1980s. Data collected using these methods are the primary tool by which the Watershed Management Division assesses the biological condition of macroinvertebrate communities. While samples may be collected at any time of year for various purposes, only samples collected during the index period from approximately September 1 – October 15 can be assessed using VTDEC macroinvertebrate biocriteria. The assessment methodology used to interpret this community data can be found in the Vermont Water Quality Standards (VTDEC, 2017).

Equipment for Lotic Semi-Quantitative Benthic Surveys:

- Kick-net - 500-micron mesh, rectangular frame 45 cm wide x 23 cm high x 25 cm deep
- Quart size containers (wide mouth preferred)
- 80% ethyl alcohol (i.e. ethanol or ETOH)
- Paper field sheets (See Figure A.1), or an electronic tablet with digital field sheets (as described in Section 3.6)
- Waders

6.5.1.1 Riffle Kick-Net Sampling

Riffle kick-net samples are used to represent the macroinvertebrate community of riffle habitats within a stream reach. Riffles are hard-bottom areas of the stream characterized by shallow depths (< 1 m) and fast, turbulent water (> 0.2 feet per second). Due to their high productivity, riffles are the best stream habitat for providing comparable data over time and across stream reaches. This method should always be used for semi-quantitative benthic surveys in high and medium gradient streams where the reach has sufficient riffle habitat for sampling. Kick-net samples represent a composite of four subsamples taken throughout the reach. The length of reach used to collect a kick-net sample should be sufficient to capture representative conditions found within the riffles of that section of stream (e.g., shading, depth, flow velocity, and substrate composition), typically not less than 25 meters.



Procedure:

1. Begin sampling in the farthest downstream section of the stream reach and work upstream. Each of the four subsample composite locations should be chosen to represent the diversity of riffle habitat conditions (e.g., shading, depth, flow velocity, substrate composition) within the stream reach being sampled.
2. Place the net on the stream bottom in a representative riffle location with the 45 cm edge perpendicular to the flow. A representative location has similar characteristics to

the overall riffle habitat present in the reach. Make sure water is flowing freely through the net and move substrate immediately downstream of the net if necessary to improve flow. Avoid artificial riffle habitat such as riprap.

3. Collect each composite subsample from an estimated 45 cm x 45 cm (0.20 m²) square area immediately upstream of the net. Move all large coarse gravel and cobble substrates to the mouth of the net and rub clean of attached organisms. Discard cleaned substrate to the side of the sample area. Portions of larger cobbles and small boulders in the 0.20 m² area that are immobile are left in place and rubbed clean of organisms with the net positioned to capture the organisms. On rare occasions in very small streams, the riffle may not have sufficient width to collect a representative composite subsample. In this case, the net may be rotated so that the 23 cm edge is perpendicular to flow. Collect the composite from an estimated 23 cm (one width) x 92 cm (four widths) rectangular area immediately upstream of the net. This will also approximate a 0.20 m² subsample area.
4. Disturb all remaining small substrate by hand to a depth of 5–10 cm and allow disturbed organic matter to flow into the net.
5. This entire riffle kick-net procedure should last a minimum of 30 seconds per composite but should continue until all substrates within the subsample area have sufficiently been cleaned and disturbed.
6. The procedure is repeated at four different riffle areas within the reach, and composite subsamples are combined into a single sample for that stream reach. The final kick-net sample will equal approximately 0.80 m² of riffle habitat.
7. After the four composite subsamples have been collected into the kick-net, large pieces of organic matter (i.e. leaves and sticks) and substrate within the net can be carefully rinsed and rubbed clean of organisms and discarded. Transfer the contents of the net into a quart sized container. Any remaining organisms attached to the net should be removed by hand (using forceps if necessary) and placed in the container. Preserve the contents of the container with 80% ethyl alcohol, submerging all collected matter with alcohol.
8. After sample collection, the kick-net should be turned inside-out and vigorously swept through the water to ensure that all macroinvertebrates have been removed.
9. A replicate sample may be needed at some sites, requiring the sampler to repeat steps 1-5. While collecting the replicate sample, it is imperative to mark and avoid areas previously disturbed with a small cairn or similar structure.
10. After completing sample collection, the sampler should make note of a general trophic rating on a scale of 0-5 (Table 6.5.A). Trophy is defined as the total weight of living biological material (*biomass*) in a river or stream at a specific location and time. This material can be observed and includes benthic algae or periphyton, vascular plants or macrophytes, benthic macroinvertebrates and fine particulate organic matter (not leaf packs, or sand) from the breakdown of all the above. The table below will help guide the observer in making the trophic rating.

Table 6.5.A Trophic rating scale descriptions for riffle kick-net macroinvertebrate samples

Trophic rating	Description
0	Almost no algae present either macro or micro algae (cannot even draw a line on substrate), cobble squeaky clean, but moss may be present. Fine organic matter also not coating surface of substrate. Macroinvertebrates very low in abundance.
1	Almost no macro algae present, micro algae light (often golden brown), can just draw a line (but no thickness to it). Moss maybe present. Fine organic matter also not coating surface of substrate. Macroinvertebrates low in abundance. Hydropsychidae caddisfly not dominant.
2	Scattered macro algae present, micro algae mostly golden brown with noticeable thickness (up to 1mm), moss can be present, and lush. Fine organic matter can be present coating surface of substrate again very thin layer. Silt rating always 2 or less. Macroinvertebrates moderate in abundance Hydropsychidae caddisfly not dominant but noticeable.
3	Macro algae more common, filaments generally less the 3", and noticeable in favored microhabitat. Micro algae can be up to 2mm thick and appear more brownish, blue green, or green. Moss often sparse. Macrophytes present in favored microhabitats. Fine organic matter noticeable when substrate is disturbed and in back waters. Macroinvertebrates moderate to high in abundance, Trichoptera Hydropsychidae, Ephemeroptera Baetidae and Ephemerellidae abundant.
4	Macro algae often dominant, filaments mostly over 3" in length. Micro algae a thick coating 2-3mm, brownish, blue green, or green colored. Macrophytes abundant in favored habitats. Accumulation of fine organic material present very noticeable 1-3mm thick, Hydropsychidae can be highly dominant, clinging to kick nets, Chironomidae noticeable in field.
5	Macro algae can be lush and often over 6" in length. Micro algae can be very thick up to 5mm brown or blue green. In extreme cases sewage fungus present. Macrophytes can be abundant in favored habitats, often coated with algae or fungus. Organic material abundant in all habitats when substrate disturbed can smell of sulfur (rotten eggs). Macroinvertebrates can be either very abundant or scarce with Diptera, Isopoda, Oligochaeta. The only EPT group noticeable is Hydropsychidae if present.

6.5.1.2 Multi-Habitat Sweep-Net Sampling



The sweep-net sampling method is used in wadeable low gradient streams, which are generally characterized by a reach having a majority soft/fine sediment, where the water velocity is slow (< 0.2 feet per second), and where the surface flow is generally smooth and unbroken. Because riffles are absent from the reach (or are small, rare, and non-representative of reach conditions), the macroinvertebrate community is sampled from “best available habitats”. Productive macroinvertebrate habitats in these streams typically consist of woody debris

snags, exposed underwater root systems, overhanging and submerged riparian woody or herbaceous vegetation, and/or macrophytes. Sweep-net samples are collected using the same sampling net used for riffle kick-net samples, and also represent a composite of four subsamples taken throughout the reach. The length of reach used to collect a sweep-net sample should be sufficient to capture an adequate quantity and diversity of low gradient habitats that are representative of that section of stream, typically not less than 25 meters.

Procedure:

1. Survey the reach visually for macroinvertebrate habitat from the shoreline before sampling begins. Note the type, abundance, and relative proportion of macroinvertebrate habitat. Each of the four composite subsamples will be taken from a single habitat type. To the extent possible, capture the diversity of habitats available, and in the relative proportion those habitats are found within the reach in the four composites. For example, in a stream reach with macroinvertebrate habitat primarily dominated by overhanging, submerged branches with some macrophytes present, the sampler may apportion those habitats 3:1 among the four composites.
2. Begin sampling in the farthest downstream section of the stream reach and work upstream. Collect each of the four composite subsamples in a downstream to upstream direction.
3. Each composite subsample is collected by disturbing the habitat and collecting the dislodged organisms in the net. This is typically done by jabbing the net into the habitat from a downstream direction and pulling back rapidly to dislodge animals, then sweeping through the same area again to capture dislodged animals. Repeat this jabbing and sweeping motion several times at each composite location. For habitats that may be relatively immobile (e.g. woody debris snags) and/or in areas with slightly faster flow, it may be necessary to rub the habitat clean by hand and sweep the net through the area adjacent to or downstream of the habitat as the dislodged organisms are displaced by the flow.
4. To the extent possible, the area disturbed for each composite should approximate a 45 cm x 45 cm (0.20 m^2) or similar surface area of stream habitat.
5. Do not sample substrate on the stream bank or stream bottom.
6. After the composites have been collected into a single sweep-net sample, rinse large pieces of organic matter within the net and rub clean of organisms before discarding.

Transfer the contents of the net into a quart sized jar. Remove any remaining organisms attached to net by hand and place in the jar. Preserve the contents of the jar by submerging all collected matter with 80% ethyl alcohol.

7. Indicate the habitat type of each of the four composites on the field form, as well as a qualitative assessment of general habitat quality and quantity throughout the stream reach (e.g. Poor, Fair, Good, Very Good, Excellent).
8. After sample collection, turn the kick-net inside-out and vigorously swept through the water to ensure that all macroinvertebrates have been removed.
9. A replicate sample may be needed at some sites, requiring the sampler to repeat steps 1-6. While collecting the replicate sample, it is imperative to mark and/or avoid areas previously disturbed.

6.5.2 Lotic Quantitative Benthic Surveys

6.5.2.1 Artificial Substrate (Rock-filled baskets)

While the semi-quantitative kick-net methodology described in Section 6.5.1 provides a good representation of the naturally established macroinvertebrate community dwelling among the substrate of a given riffle, the use of artificial substrates can decrease variability associated with habitat and substrate within and between stream sites. The colonization of artificial substrates by macroinvertebrates over a given period provides quantitative method of replication by controlling for this habitat variability while still reflecting changes in water quality due to other ambient factors.

Rock-filled basket artificial substrates are the preferred method for quantitative, replicated macroinvertebrate collections. This methodology is appropriate for medium to high gradient, hard-bottomed stream reaches, and is not applicable for large rivers or low gradient streams, or other aquatic ecosystems such as lakes, wetlands. This method was specifically developed by the VTDEC for use in paired site comparisons for determinations of compliance with the Aquatic Biota Criteria of the 2019 Indirect Discharge Rules, as revised (VTDEC 2019). Deployment of rock-filled baskets for macroinvertebrate colonization is generally a five-to-six-week period. Deployments may occur during spring or summer if specified for a given study but will typically occur during the months of August – October. Collection of the artificial substrates should occur during the index period for semi-quantitative kick-net sampling (approximately September 1 – October 15) to allow for sample collection using both methods. While this method has been used historically to bracket impact areas and spatial controls at indirect discharge sites, there may be additional utility for other types of studies.

Equipment:

- Rock-filled basket artificial substrates (Figure 6.5.2.A.) – 8/site. Constructed using:
 - Rectangular basket made of ½" hardwire cloth, with dimensions 20 cm x 30 cm x 5 cm (7.5" x 12" x 2"),
 - Filled with 10 lbs. of 1 ¾" (4.5 cm) graded granite rock
- Quart size containers (wide mouth preferred)
- 80% ethyl alcohol (i.e., ethanol or ETOH)
- Sieve bucket – mesh size 500-595 um
- Meter stick or depth tape
- Measuring tape
- Handheld scrub brush
- Paper field sheets, or an electronic tablet with digital field sheets
- Waders

Procedure:

1. Install the artificial substrates on the streambed, positioned such that the long axis is parallel to flow.
 - a. Choose installation locations for similarity of microhabitat, including flow velocity and depth. When using these methods for paired site comparisons, similarity of

- microhabitat must also be considered and maximized between the paired stream reaches.
- b. Place artificial substrates in riffles or runs (erosional areas) comprised of coarse substrates. Installation locations should be in or near the thalweg, or other areas with sustained moderate flow (i.e., not in depositional areas like pools or backwater). To the maximum extent possible, place artificial substrates on the streambed consistently between baskets at a given site and between paired sites.
 - c. Place artificial substrates directly on top of the surface of the streambed to decrease the likelihood of being mobilized during a freshet flow event. This will require the clearing of coarse native substrate from the installation location, though the artificial substrate should not be further worked into the streambed. Aim to place the artificial substrate at approximately the same level as adjacent native substrate.
 - d. Place artificial substrates consecutively and longitudinally in the channel, typically spaced at least 3' apart to minimize potential influences in flow dynamics between substrates.
2. Record relative position in the channel, depth at the top surface, and estimated velocity of the water at the upstream edge for each installed artificial substrate. Relative position in the channel can include a sketch of substrate locations and/or measurements taken from a single fixed location adjacent to the channel to aid in locating substrates for retrieval. Starting upstream, number substrates one through eight for collection purposes.
 - a. Artificial substrates are left in the channel for a period of 5-6 weeks and then collected. If high flows are initiated by a single substantial precipitation event (e.g., ≥ 2 " rain) or several significant smaller events near the end of the study period, delay the collection of substrates until at least one week after high flows have receded.
 3. Prior to removal, record the relative position in the channel, depth at the top surface, and estimated velocity of the water at the upstream edge for each artificial substrate. Assign each substrate a condition rating of 1-4 using the scale below. Substrates rated as 1 or 2 will be selected for the study, starting with the lowest numbered substrates. If there are not enough substrates rated 1-2, a substrate with a rating of 3 may be used. Substrates with a rating of 4 will be rejected from further use in the study.
 - a. 1 – Undisturbed
 - b. 2 – Minimal sediment, embeddedness, and/or movement. Depth, velocity and orientation maintained.
 - c. 3 – Some sediment, embeddedness and/or movement. Depth, velocity and orientation altered, but substrate remains fully submerged with noticeable water movement.
 - d. 4 – Significantly dislodged, was not submerged or lacked noticeable water movement, and/or had very poor orientation or was buried.
 4. A total of six artificial substrates will be collected at each site, with lowest numbered substrates of sufficient quality chosen to eliminate bias. A minimum of five substrates at each site are required for subsequent lab processing and data analysis, with the sixth substrate retained as an additional replicate. If there are not five substrates of sufficient quality for collection at each site (based on the individual substrate ratings and the professional opinion of the field biologist), the artificial substrate study must be abandoned. Macroinvertebrates will then be collected at each site using the grid method in Section 6.5.2.2.

5. Carefully remove any loosely accumulated debris such as leaves or vegetation from the outside of the artificial substrate. Debris that has become attached or incorporated into the artificial substrate should be left intact and collected as part of the sample.
6. While carefully removing each artificial substrate from the streambed, hold the sieve bucket against the downstream edge of the substrate to collect any macroinvertebrates dislodged. Open the artificial substrates and empty the contents into the sieve bucket (Figure 6.5.2.B.).
7. Wash, rinse, inspect, and set aside each piece of granite from the artificial substrate until the bucket only contains macroinvertebrates, other organic detritus and finer sediment.
8. Carefully transfer the contents of the bucket to a quart-sized container and preserve in 80% ethyl alcohol for subsequent laboratory processing. For typical studies (i.e., indirect discharge), five of the six replicates will be processed in the lab for further analysis. Retain the additional replicate as a replacement if one of the five primary replicates is lost or compromised.



Figure 6.5.2.A Photo of a rock-filled basket artificial substrate



Figure 6.5.2.B Emptying contents of rock basket into sieve bucket

6.5.2.2 Riffle Kick-Net with PVC Grid

This method is meant to provide a quantitative substitute for the artificial substrate rock-filled baskets described in 6.5.2.1. Similar to the artificial substrates, this method was specifically developed by the VTDEC for use in paired site comparisons for determinations of compliance with the Aquatic Biota Criteria of the 2019 Indirect Discharge Rules, as revised (VTDEC 2019). This method involves the placement of a rectangular PVC 'grid' directly on the riffle substrate, and the subsequent disturbance and collection of macroinvertebrates from that defined area. It is designed to be replicated numerous times at each site to minimize natural variability to the extent possible. It is appropriate for riffle areas in high or medium gradient wadeable streams.

The PVC grid method is an acceptable substitute only when the use of artificial substrates is not feasible, such as in streams where the size, habitat, or flow characteristics of the stream make the proper deployment of artificial substrates impossible. This would most likely occur in a small shallow stream where a series of rock-filled baskets cannot be fully submerged and

therefore the macroinvertebrate colonization would not be replicable and representative of stream conditions. This condition would be determined before a study began, and in the case of indirect discharge compliance monitoring, the applicability of this method would be confirmed by a VTDEC biologist. The PVC grid method may also be used as a substitute when a high flow event has disrupted the artificial substrates during the exposure period, causing a significant number of artificial substrates to be dislodged, buried in sediment, or otherwise compromised. If five replicate artificial substrates of sufficient quality for macroinvertebrate sampling are not able to be collected from both sites of a paired site comparison, the PVC grid method will be used instead.

Equipment:

- Weighted rectangular PVC 'grid', interior opening size 45 cm x 30 cm (18" x 12")
- Kick-net - 500-micron mesh, rectangular frame 45 cm wide x 23 cm high x 25 cm deep
- Quart size containers (wide mouth preferred)
- 80% ethyl alcohol (i.e. ethanol or ETOH)
- Paper field sheets, or an electronic tablet with digital field sheets
- Waders

Procedure:

1. Visually inspect the study area before the collection of replicates using the PVC grid. Choose the six riffle locations for similarity of microhabitat, including substrate size distribution, flow velocity, and depth. The natural substrate at each collection location must be of a size that will allow the grid to be placed directly on the streambed (i.e., large cobbles and boulders will not be suitable). As these methods are typically used for paired site comparisons (i.e., indirect discharge studies), similarity of microhabitat must also be considered and maximized between the paired stream reaches.
2. Conduct PVC grid collections in a downstream to upstream direction within a site to minimize potential effects of disturbance on subsequent collection areas.
3. At each riffle collection location, the grid is placed directly on the streambed with the long axis perpendicular to flow. Adjust native substrate carefully to allow the grid to sit directly on the streambed. Place the rectangular kick-net immediately downstream of the grid to collect dislodged organisms.
4. Move all large coarse gravel and cobble substrates inside the grid to the mouth of the net and rub clean of attached organisms. Discard cleaned substrate to the side of the sample area. Portions of larger substrates inside the grid area that are immobile are left in place and rubbed clean of organisms.
5. Disturb all remaining smaller substrate by hand to a 5–10 cm depth and allow disturbed organic matter to flow into the net.
6. At each collection location, carefully transfer the contents of the net to a quart-sized container and preserve in 80% ethyl alcohol for subsequent laboratory processing. For typical studies (i.e., indirect discharge), five of the six replicates will be processed in the lab for further analysis. The additional replicate should be retained as a replacement if one of the five primary replicates is lost or compromised.

6.5.3 Lentic Qualitative Littoral Zone Benthic Survey

The lake littoral zone is made up of many microhabitat types that have a strong influence on the macroinvertebrate species present. To qualitatively inventory the macroinvertebrate species of a lake the following microhabitats should be sampled in a lake:

- a. Rocky/Cobble/Large Woody Debris
- b. Macrophyte Beds
- c. Fine organic material and leaves

Equipment:

- Kick-net - 500-micron mesh, rectangular frame 45 cm wide x 23 cm high x 25 cm deep
- Forceps, strainer
- Sieve bucket – mesh size 500-595 um
- Quart size jars (wide mouth preferred)
- 80% ethyl alcohol (ethanol or ETOH)
- Field sheets
- Waders

Procedure:

1. Record Physical Characteristics of sampling site (see Section 3.8 Physical Habitat Characterization - Lentic)
2. Collect animals from all habitat types by sweeping with kick-net through macrophytes, mud and leaf litter. Rinse off woody debris, macrophytes and rocks through a kick-net or sieve bucket. Pick animals off substrate of large wood and cobble.
3. Place collected animals and rinsed material in wide mouth jar and cover with 80% ethanol.
4. Label jar with lake name and date.
5. Record location of habitat sampled on field sheet or map.

6.5.4 Lentic Semi-Quantitative Benthic Survey

To evaluate the overall macroinvertebrate biological integrity of Vermont lakes when collecting samples from the littoral and sublittoral zones, the lake should be trisected into equal areas of shoreline. Samples will then be collected from within each trisection of the lake and composited to represent the overall condition of the lake.

The lake littoral zone is made up of many microhabitat types that have a strong influence on the macroinvertebrate species composition present. Therefore, the littoral zone macroinvertebrate assessment protocols will stratify sample collection based on specific habitat types. Qualitative sampling of each habitat type will take place at three random sites on a lake. Samples of each habitat type will be composites from the three sites to represent the macroinvertebrate assemblage of the lake littoral zone for each microhabitat type. The following microhabitats will be assessed separately:

- a. Rocky/Cobble/Large Woody Debris
- b. Macrophyte Beds
- c. Fine organic material and leaves

Equipment:

- Kick-net - 500-micron mesh, rectangular frame 45 cm wide x 23 cm high x 25 cm deep
- Ekman dredge, 6"x 6" square, (15 x 15 cm)
- Quart size jars (wide mouth preferred)
- 80% ethyl alcohol (ethanol or ETOH)
- Sieve bucket – mesh size 500-595 um
- Field sheets
- Waders

Procedure:

1. Record physical characteristics of each sampling site. See Figure A.5. Lentic Physical Habitat Assessment Form in the Appendix.
2. Rocky/Cobble/Woody Debris
 - a. Composite three sites with 10 minutes searched per site.
 - b. Pick up and wash off rocks and woody debris through a kick net or sieve bucket and pick off animals from substrate with forceps.
 - c. Place representative animals in quart size jars.
 - d. Cover with 80% ethanol and label jar with date, lake name and habitat sampled.
3. Macrophyte Beds
 - a. Composite three sites with 3 sweeps per site.
 - b. Sweep kick-net through representative macrophyte beds. Do not sample the lake bottom.
 - c. Place representative animals in quart size jars.
 - d. Cover with 80% ethanol and label jar with date, lake name and habitat sampled.
4. Muddy habitat- fine organic material and leaves
 - a. Composite three sites with 3 kick-net sweeps per site.
 - b. A sweep kick-net is run through the top 5 cm (2.0") of bottom muck.
 - c. Sieve mud through kick net or sieve bucket.
 - d. Place sample into quart size jar.
 - e. Cover with 80% ethanol and label jar with date, lake name and habitat sampled.

6.5.5 Lentic Quantitative Benthic Survey

6.5.5.1 Lake Rocky/Cobble Littoral Zone

Quantitative sampling of the benthic community within the lake rocky/cobble littoral zone is conducted by use of SCUBA quadrat areal sampling methods.

Equipment:

- one-quarter square meter quadrat
- Large 500 micron mesh bag
- Quart size jars (wide mouth preferred)
- 80% ethyl alcohol (ethanol or ETOH)
- Sieve bucket, No. 30, mesh size 500-595 μm
- Field sheets

Procedure:

1. Remove all substrate within the one-quarter square meter quadrat and place into a large 500 micron mesh bag.
2. Seal the bag and return to the surface (boat/shore) for further processing.
3. Wash each sample through a U.S. Standard No. 30 sieve bucket (600 micron).
4. Place the retained sediment, debris, and organisms into labeled jars and preserve with 80% ethanol.

6.5.5.2 Lake Profundal Zone (muck, soft bottom)

Equipment:

- Ekman dredge, 6"x 6" square, (15 x 15 cm)
- Measuring Tape
- Quart size jars (wide mouth preferred)
- 80% ethyl alcohol (ethanol or ETOH)
- Sieve bucket, No. 30, mesh size 500-595 μm
- Squirt bottle
- Field sheet and lake map
- Hydrolab
- Kemmerer
- DO sample bottles and preservation
- Secchi disk
- 0.5 m^2 quadrat



Procedure:

1. Prior to collecting sample, record physical characteristics of sampling site (Section 3.8 Physical Habitat Characterization - Lentic).
2. Identify sample site location from 3 shoreline markers, and/or lake map.
3. Measure water depth, temperature profile. Collect dissolved oxygen sample 1 meter above bottom.
4. Record the following (if applicable):
 - a. Secchi depth
 - b. Ice thickness, and snow cover
 - c. Date, time, water temperature

- d. Sampling crew names
- e. Weather, air temperature, wind
- f. Substrate composition of dredge sample
- g. Equipment used, other sampling conducted
5. Sampling
 - a. Lower Ekman dredge to within 2 meters (6.5 ft) of bottom.
 - b. Allow dredge to stabilize (hang straight down, stop swinging).
 - c. Allow dredge to drop into bottom muck.
 - d. Release messenger to dredge.
 - e. Retrieve dredge with a smooth steady pulling action.
 - f. Place dredge into No. 30 sieve bucket and empty contents.
 - g. Record substrate type and composition.
 - h. Sieve out fine muck by dipping bucket onto water surface with a rotating motion.
 - i. Pour remaining sample into quart jar.
 - j. Use squirt bottle with ethanol to clean remaining sample from No. 30 sieve bucket into quart jar.
 - k. Cover sample with 80% ethanol and label jar with lake name, lake zone, date.

6.5.6 Lentic Littoral Habitat Macroinvertebrate Collection Methods

As part of the Littoral Habitat Assessment, macroinvertebrates are collected at rocky littoral and sandy littoral sites using different methods depending on the substrate type.

Rocky Littoral Sampling

This sampling employs the same method as the Lentic Quantitative Benthic Survey (LQBS). Samples are collected at the 0.5 m depth contour in the Littoral Habitat Plots. Three 1 m² quadrats are laid along the transect where the most cobble is present. Follow the same procedure for filling the 500 um mesh bags as outlined in the LQBS.

Equipment:

- 6.5 cm diameter core tubes (5) with caps
- Quart size jars (wide mouth preferred)
- 80% ethyl alcohol (ethanol or ETOH)
- No. 30 mesh sieve bucket
- Field sheet and lake map

Five 6.5 cm diameter core tubes are driven 10 cm into the sediment along the 0.5 m contour and Littoral Habitat shallow water transect. They are then capped and carefully extruded from the sediment being careful not to lose any of the sediment by cupping your hand under the tube as it emerges from the sediment. All cores are then washed through a U.S. Standard No. 30 sieve (600 micron). Place the retained sediment, debris, and organisms into labeled jars and preserve with 80% ethanol.



Figure 6.5.6.A. Sandy littoral sampling

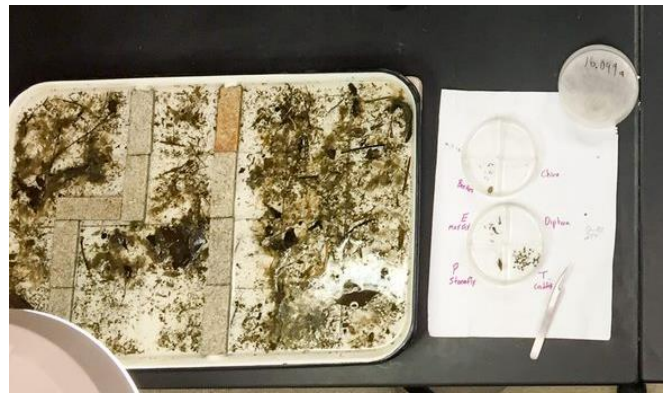
6.5.7 Macroinvertebrate Laboratory Sample Processing Methods

The following method describes how all semi-quantitative and quantitative macroinvertebrate samples are processed (including standardized sub-sampling methodology) for estimates of density, taxa richness, and other community metrics. Sub-samples are initially processed by removing organisms from organic and inorganic detritus under 2x magnification and sorting by taxonomic order. The sub-sample is fully processed (picked) by one individual and is then checked by a second qualified technician or biologist who has been certified to conduct secondary checks of picked samples. The second individual checks through the picked portion of the sample (either the sub-sample or whole sample) for completeness, removing any remaining organisms.

6.5.7.1 Macroinvertebrate Sample - Quantitative Processing

Equipment:

- 80% ethyl alcohol (ethanol or ETOH)
- No. 30 standard test sieve (595 um)
- 12" x 18" white (enamel or plastic) tray, surface of tray delineated into 24 equal squares
- 3" - 4" fine pointed watchmaker forceps
- Phloxine B red dye (optional)
- 2-5 gallon plastic bucket
- 1 inch x 1–3-inch x ¼ inch blocks (made of granite or similar material)–
- Hand counter
- Quarter sectioned plastic petri dishes
- Wax pencil for labeling petri dishes
- Tap water
- 2x magnification illuminator



General Processing Procedure:

1. Position a No.30 sieve over a bucket. Open sample container and pour contents into the sieve, allowing the ethanol to collect in the bucket. Discard ethanol in a labeled waste receptacle.
2. Rinse contents of sample container into the sieve using a hand-held sink sprayer. Rinse sample thoroughly.
3. Gently rinse large pieces of organic matter and inorganic substrate in the sieve and then discard.

4. Backwash sample from sieve into delineated tray.
5. Spread sample evenly over entire tray surface. Add enough water to aid in spreading the sample evenly. Note: Water should just cover gravel and debris; too much water will cause animals to float around tray (~ 1/3-1/2" deep), or affect ability to sub-sample as necessary.
6. For full sample processing, pick all macroinvertebrates from tray and sort into major taxonomic groups (orders) in petri dish quadrants filled with 80% ethanol. After five minutes of additional searching, if no animals are found the sample is considered processed by initial picker. The sample is then checked by the qualified technician/biologist before initial processing is complete.



Sub-sampling Processing Procedure:

For semi-quantitative lotic macroinvertebrate sampling and assessments, as described in Section 6.5.1 and in Appendix G of the Vermont Water Quality Standards (VTDEC 2017), a minimum 25% subsample is required to apply biological criteria. If the 25% subsample does not yield 300 organisms, the subsample is incrementally increased until a total 300 organisms is reached, or the full sample is processed. The following is the protocol for this subsampling procedure.

1. Follow steps 1-5 of the general processing procedure (above).
2. After sample is spread evenly onto tray, use a random number generator to produce a random number between 1 and 24. Start at this number and block off a total of 6 consecutively numbered squares using the granite blocks (or similar suitable device). Gaps between the blocks and the tray may be filled with a small piece of rolled up paper tissue to prevent animals from floating in or out of the picked area during processing.
3. Pick all organisms from this initial six-square subsample and sort by taxonomic group (order) into petri dish quadrants filled with ETOH. Label petri dish covers with the laboratory ID number using a wax pencil.
4. Count the number of organisms removed from the initial sub-sample using a hand counter. If less than 300 organisms are removed from the first six squares, estimate the number of additional squares needed to reach the 300 organism minimum. Adjust the granite blocks to include the expanded subsample. Fully pick the additional squares in the expanded sub-sample. Once a square is added to the sub-sample area, all organisms from that square must be removed. If the expanded subsample does not result in the minimum of 300 organisms, the subsample is again expanded. This may result in the processing of the entire tray if the density is very low. Record the final number of squares processed, who processed the sample, and the date of processing on a laboratory bench sheet.
5. The final subsample is then checked by a qualified technician or biologist, who attempts to remove any remaining organisms into the petri dishes from the tray before disposal of the sample. This biologist may scan the unprocessed portion of the sample for any large and/or rare taxa that may not have been found in the processed subsample. These large/rare organisms must be kept in a separate labelled "L&R" petri dish, and not quantitatively used as part of the final data set. The identification of these organisms is

for distributional purposes only. Record the initials of the sample checker on the laboratory bench sheet.

6. Ten percent of samples processed annually require a third individual (qualified technician or biologist) to check the sample and tally any remaining organisms found in accordance with the Biomonitoring Program Quality Assurance Project Plan (VTDEC 2018). The number of organisms found in this additional check is recorded on the laboratory bench sheet and the additional organisms are added to the petri dishes.

Variations on sample processing:

A. “Swirling” technique

1. Samples can be gently “swirled” to separate the organic matter (including the macroinvertebrates) from sand and gravel. This technique significantly improves processing ease and efficiency when there is a high proportion of sand and gravel in the sample jar. It is important to swirl the sample gently to avoid damaging fragile macroinvertebrate body parts such as mayfly gills.
2. Decant the ethyl alcohol through the No. 30 standard test sieve into a waste container, and then rinse the sample into the sieve. Rinse and remove large pieces of organic and inorganic matter in the sieve and then discard. Transfer the remaining sample from the sieve into a 2–5-gallon bucket. Fill the bucket ~25% with water. Gently swirl the contents of the bucket. The lighter materials, including the animals, will float in the water. Pour off the water and floating organic material into the sieve, leaving the sand and gravel in the bucket. Repeat these steps several times, until all organic material appears to be removed from the bucket.
3. When the sample has been successfully swirled, set the bucket with the remaining sand and gravel aside with a sample label from the quart jar to be processed later. The sand and gravel will need to be checked for organisms when the primary sample has been completed. Stone-cased caddisflies and other animals could remain in the separated sand and gravel. Subsample this material the same way that the primary sample is processed, being sure to remove organisms from the exact same squares in the 24-grid tray.

B. Phloxine B Red Dye.

1. The sample can be dyed to aid in seeing and removing the smaller animals (especially small worms and dipterans). This may especially be necessary in samples with a large amount of non-animal organic detritus (e.g. semi-quantitative lotic multi-habitat samples containing abundant plant material).
2. Review the Phloxine B Material Safety Data Sheet and use necessary personal protective equipment.
3. Dip a long (~10 cm in length) metal spatula into the Phloxine B bottle to remove a small amount of dye that covers the tip of the spatula by 1-2mm.
4. Open the sample container and insert the spatula to add the dye to the sample.
2. Gently invert jar to dissolve and distribute dye. For densely packed samples, the dye will need to be spread throughout using the spatula.
3. Allow sample to sit for 24 hours before processing.

6.5.7.2 General Taxonomic Identification and Enumeration:

Identification and enumeration of organisms from the processed sample are used to calculate community metrics, which are used to assess the condition of the macroinvertebrate community using the State's biological criteria.

Equipment:

- 80% ethyl alcohol (ethanol or ETOH)
- Binocular dissecting microscope 7x - 60x minimum range
- Fiber optic illuminator
- Petri dishes –quartered
- VTDEC listed taxonomy keys
- Acid free labeling paper printed with fade-resistant indelible permanent ink (Not a Sharpie)
- 3-4" fine pointed watchmakers forceps
- Fine pointed probes
- Scalpel - #15 blades
- 20 - 30 ml snap cap glass vials
- ¼ dram - 2 dram open glass vials
- Cotton
- 500 ml screw cap, wide mouth, glass jars

Procedure:

1. After a sample has been initially processed and sorted by taxonomic group into petri dishes under 2X magnification, conduct a more thorough and accurate sort under higher magnification. Sort animals by taxonomic groups (order) using a dissecting microscope and place in snap cap vials with 80% ethanol and labels indicating sample laboratory ID number.
2. Identify groups to lowest possible or recommended taxonomic unit (usually genus or species) using a binocular dissecting microscope and keys recommended VTDEC.
3. Identification of certain groups (e.g., Chironomidae, Oligochaeta) or individuals may require slide mounts for identification with a compound microscope (see method 6.5.7.3).
4. Organism identifications and number of individuals in each taxonomic unit are recorded by the taxonomist and added to a VTDEC Microsoft Excel template for that specific laboratory ID number. Each row for a specific taxa must include the unique VTDEC macroinvertebrate "key number" that will allow that taxa to be uploaded to the VTDEC database. The taxonomist also records their initials with each identification and assigns a confidence rating of A (99% confident), B (90% confident) or C (75% confident). Typically, biologists will not use a "C" rating and will instead use a broader level taxonomic classification (e.g., genus instead of species).
5. Organisms are archived by taxonomic order in glass vials, in 80% ethanol, and identified by laboratory ID number.
6. Vials are capped with cotton wads and stored by year sampled and order groupings in 500-1000 mL screw cap archive quality jars topped with 80% ethanol.

6.5.7.3 Chironomidae and Oligochaeta Identification.

The following protocols are used to determine the identification of genera and species for Chironomidae and Oligochaeta organisms in a sample unit, if necessary. VTDEC assessments based on semi-quantitative lotic protocols (Section 6.5.1) typically require this level resolution for Chironomidae, however Oligochaeta usually only require identification to the family level.

Equipment:

- 80% ethyl alcohol (ethanol or ETOH)
- Mounting Media - Hoyer's Media or a low-toxicity media of similar quality
- Binocular Dissecting Microscope - 7x - 60 x minimum range
- Fiber optic illuminator
- Compound Microscope with phase contrast - 40 - 400x, 1000x oil immersion
- Divided petri dishes
- Microscope slides - 75 x 25 mm x 1 mm thick
- Cover glass slips - 22 x 22 mm, thickness 0-1
- Slide boxes
- VTDEC listed taxonomic keys
- Labeling stickers, permanent ink
- 3-4" fine pointed watchmakers forceps
- Scalpel with #15 blade

Procedure:

1. To the extent possible with a high level of confidence, presort Chironomidae and Oligochaeta into unique genus or species groups using a dissecting microscope.
2. Mount representative individuals of each grouping on individual microscope slides under a dissecting microscope. One to two drops of media is applied to the slide and lightly spread. Usually 2 - 6 organisms can be mounted per cover slip area, equaling 4-12 animals per slide. This may vary depending on the experience of the biologist and the size of the organisms. All organisms should be oriented in the same direction for mounting. Chironomidae specimens should be mounted with the ventral head surface pointed upwards.
3. The taxonomist should be familiar with the specifications of the mounting media being used and adhere to any safety recommendations provided by the manufacturer. Mounting should generally be performed in the presence of ventilation equipment (e.g. a snorkel vent).
4. Sorted genus/species groups represented by many individuals (> 10) may be subsampled in sufficient amounts to ensure a correct identification of the group (10-50% depending on the distinctiveness of the group). Typically, not less than 5 individuals or 10% (whichever is greater) will be identified from a subsampled genus/species group. This may vary based on the sorting experience of the biologist. If all organisms subsampled are identical, the total enumeration (including those not mounted) is recorded. If more than one species is found in the mounted subsample, the ratio of species to the unmounted organisms must be determined.
5. Labels are marked to identify which sample the organisms came from using the laboratory ID number.

6. Organisms are identified under a compound scope using keys recommended by VTDEC. Genus/species identifications and number of individuals of each taxonomic unit are recorded by the biologist and added to a VTDEC Microsoft Excel template for that specific laboratory ID number. Each row for a specific taxa must include the unique VTDEC macroinvertebrate “key number” that will allow that taxa to be uploaded to the VTDEC database. The taxonomist also records their initials with each identification and assigns a confidence rating, ranging of A (99% confident), B (90% confident) or C (75% confident). Typically, biologists will not use a “C” rating and will instead use a broader taxonomic classification (e.g.,. genus instead of species).

6.6 Mussels

Of Vermont's 18 native species of freshwater mussels, 10 species (55%) are listed as threatened or endangered ([Vermont Atlas of Life, 2017](#)). Native freshwater mussel populations must contend with habitat loss, water level fluctuations, invasive zebra mussels, and changing thermal regimes due to climate change. Like other macroinvertebrates, mussels are water quality indicators and are sensitive to pollution and disturbance. As such, they are as vulnerable as they are a critical component of the aquatic ecosystem. Surveys of freshwater mussel status and trends is a component of VTDEC biomonitoring.

Equipment:

- Mesh bags for collecting mussels
- Sieve buckets for holding mussels
- Calipers to measure mussel length, width, and height
- Stopwatch
- Clipboard with field sheets on waterproof paper
- Pencils, Sharpie, labels
- Plastic sealable bags for empty shells
- Snorkel, mask and wetsuit for each data collector
- Viewing buckets
- SCUBA equipment for lentic surveys occurring at depth
- Underwater writing pads for each data collector
- Cameras that can take photographs underwater
- Secchi disk
- Depth rod (2-meter)
- Measuring tape (50-meter or 100-meter) for line transects
- 1-meter PVC square for transects
- Pin flags for marking the locations of rare, threatened, or endangered individuals

6.6.1 Shallow Depth Mussel Surveys

For waters with an average depth of 1-2 meters or less, it is possible to conduct semi-quantitative and quantitative mussel surveys using a mask and snorkel. Crews of at least 3 people are ideal, where one crew member is a data recorder and the other two are the data collectors/observers.

1. In semi-quantitative surveys, collectors/observers snorkel and search for mussels for a set period of time. During the search time, the observers search for mussels in the substrate and collect individuals in a mesh bag.
2. Mussels should be removed slowly from substrate to avoid damaging their foot. Locations of rare, threatened, or endangered species should be marked with a pin flag so that they can be returned.
3. The data recorder keeps track of time and alerts the observers when time is up. After the timed search is over, the mussels are transferred to a sieve bucket filled with water. To minimize mussel stress, time out of water should be minimized. Live mussels are not killed or collected during surveys, though empty shells are collected and labeled with the survey site location.

4. Mussels are removed one at a time from the sieve bucket and identified. Catch Per Unit Effort (CPUE) is calculated by dividing the number of mussels found by the total number of person-hours spent on the timed search. For example, 2 people searching for 30 minutes would equal 1-person hour. Ten mussels found during this time period would equal a CPUE of 10 mussels/hour. Take photographs of each species present for documentation.
5. Measure shell length, width, and height using calipers. Note shell condition based on percentage of periostracum erosion (<10% = Light, <20% = Light-Medium, <30% = Medium, <40% = Medium-Heavy, >40% = Heavy).
6. After mussels have been processed, they are returned to the substrate with their posterior (siphoning) end facing up. Note the average depth of the survey area, stream velocity and substrate characteristics at the time of sampling.
7. Alternatively, in areas where mussels are dense, conduct a timed search and use abundance categories instead of collecting individual mussels.
8. If snorkeling is not possible for whatever reason but the depth is still relatively shallow, use viewing buckets to observe mussel abundance categories or conduct qualitative surveys of mussel presence or absence. Care must be taken not to step on mussels or create turbid water conditions that make mussels difficult to see. Due to limitations of the viewing bucket, snorkeling is the preferred mussel survey method.
9. Quantitative surveys can supplement semi-quantitative surveys by using line transects of at least 10m in length or 1-m square transects. For line transects, collect and process all mussels within 5cm on either side of the line. Square transects can also be used to quantify mussels/square meter. Thoroughly search and excavate the area within each transect to 10cm to find mussels that may be buried in sediment.
10. Additional information on shallow-water mussel survey methods by snorkeling can be found in the [Brook Floater Rapid Assessment Monitoring Protocol](#) (USFW 2018).

6.6.2 Deep Water Mussel Surveys

For waters deeper than 2 meters, such as large rivers or lakes, qualitative snorkel surveys and timed searches may still occur, though increased depth presents significant challenges to semi-quantitative and quantitative data. Use SCUBA equipment to conduct mussel surveys at these depths following the procedures used in the Survey of Native Mussel Beds in Lake Champlain (VTDEC 1995).

6.7 Fish

The electrofishing sampling methods described here have been used to determine abundance and species composition of fish populations in Vermont's rivers and streams since the mid-1980s. Data collected using these methods are the primary tool by which the WSMD assesses the biological condition of a fish community. Presently, use of the VT Indices of Biological Integrity (IBIs) are restricted to wadeable streams supporting at least two native species. Selection of the sampling site is a critical step in fish community evaluation regardless of index used or program goal. The habitat of the stream reach sampled must be representative of the stream in which it is located. Fish surveys may occur at any time of year, but fish communities can only be assessed when surveyed during the index period from approximately September 1 – October 15. The field sampling and assessment methodology used to interpret this community data can be found in the Vermont Water Quality Standards, Appendix G (VTDEC, 2017).

6.7.1 Stream Electrofishing

Safety:

All staff regularly involved in electrofishing operations must successfully complete the 3-hour online course [Electrofishing Safety \(CSP2202\)](#) offered by the U.S. Fish & Wildlife Service and read and understand the VTDEC Safety Policy for Electrofishing Operations (VTDEC, 2018). A permit from the Vermont Fish and Wildlife Department is required to electroshock streams in Vermont. The brief safety outline provided below is not meant as a guide for the beginner. A trained and experienced biologist with a valid Scientific Collection Permit obtained from the Vermont Department of Fish and Wildlife must always be part of an electrofishing crew.

1. Never operate a shocker alone. Minimum crew size is 2 people, 3 is better.
2. Operator and netters must always wear insulating rubber linemen gloves to protect from high voltage.
3. At least two individuals on a crew must be certified in CPR and AED use.
4. Never operate a shocker in deep or fast water where footing is uncertain.
5. Crew should be supervised by an experienced fisheries biologist.
6. Avoid operation near livestock, domestic animals, and other people.
7. Electrofishing should proceed at a deliberate pace, avoiding erratic "fish-chasing" movements.
8. All participating personnel must wear chest waders in good condition (no leaks). Chest waders made out of neoprene material and long pants are strongly recommended in warm weather conditions, when sweating may cause electrical conductance through waders made out of more breathable material.

Equipment:

- Electrofishing backpack(s)
- Electrofishing batteries
- Anode
- Fiberglass handled nets (one per person)
- Chest waders
- 5-gallon bucket(s)

- Insulated, rubber lineman gloves
- Polarized sunglasses
- Live well
- Fish viewing chamber
- Magnifying glasses
- Taxonomic keys
- Fish population field sheets (See Figure A.3) on waterproof paper
- Fish section habitat evaluation sheet (see Figure A.4) on waterproof paper or electronic field form
- Measuring tape
- Laser rangefinder
- Fish measuring board demarcated in mm



Procedure:

1. Check all equipment before the field visit for problems which may hamper safety and effectiveness.
2. Delineate a stream reach of 75-200 meters (245-650 ft) depending on the goals of the sampling and average width of the stream. Minimum length shall be 75 meters regardless of width and increases with mean section width. Determine average wetted width based on 6-10 measurements over stream reach. Use Table 6.7.A to determine minimum reach length. Avoid walking in the stream before sampling to minimize disturbance of sediments and the fish community.
3. Use block nets to delineate the sampling reach when the river is deep or there are no natural barriers to prevent fish from escaping upstream. Fishing efficiency and resulting data quality is increased when escape is minimized.
4. Select electric current and pulse levels that are sufficient to stun fish effectively but not so strong as to cause mortality. To determine initial voltage prior to sampling, determine the stream conductivity, and consult Table 6.7.B for suggested voltage settings based on VTDEC data. Voltage can be adjusted as necessary during the beginning of the survey depending on observed capture efficiency and/or fish mortality.
5. Perform sampling in a systematic fashion. Begin at the downstream end of the sampling reach and move upstream, covering the total area of stream, including undercut banks and overhanging vegetation. For quantitative surveys, use two backpack shockers streams wider than 6 meters; at streams wider than 12 meters, use two shockers to sample each bank out 6 meters. When multiple shockers are used, operators should proceed upstream in parallel and at the same rate.
6. The same individual should operate the anode pole within successive passes at one reach and between reaches which are to be directly compared. Successful fishing is accomplished by simultaneously sweeping the anode back and forth and maintaining a steady upstream movement, being sure to effectively cover all available habitat types. The sampled section should include all forms of habitat (pool, run, and riffle) in

approximately the same proportion as observed in the stream reach. If possible, the section should include two pool/riffle cycles.

7. Each sampling crew consists of one shocker and a minimum of one netter carrying a 5-gallon bucket partially filled with stream water. The individual operating the shocker serves as the crew leader, determining pace and direction of shocking. Netters must stay downstream of the shocker and avoid stepping on the cathode. Captured fish should immediately be placed in a bucket filled at least $\frac{1}{3}$ full with stream water and carried by each person netting fish. Monitor collected fish for overall condition during the sampling. If they are having difficulty swimming or stay stunned, the voltage needs to be reduced. Adding or replacing the water in the collection bucket mid-survey will help the fish by adding more oxygenated water.
8. At the end of a pass or run, captured fish are placed in a live well. A live well is a large plastic tub with small holes that is submerged in an area with slow flow. The holes need to be small enough to prevent fish from escaping, and large enough to allow some water exchange. In long reaches, place a live well halfway through the sampling reach so that fish can intermittently be placed there. Do not shock near the live well when fish are inside. Care must be exercised when holding cold water species captive during summer months by maintaining acceptable water temperatures. This can be done by providing shade over the live well, or by identifying fish midway through the survey. If fish are identified mid-survey, they must be released downstream of a movement barrier that will ensure they are not recaptured when the survey proceeds.
9. All fish collected will be identified to species if possible and the total count will be recorded on field sheets. Only identify and record fish greater than one inch (2.5 cm). Notation will be made of presence and relative abundance of young of the year fish and any deformities or abnormalities recorded (e.g., black spot, yellow grub, fin erosion, deformities, etc.). Lengths are measured in mm for all salmonids captured to assist in determining the number of age classes present. If fish cannot be identified stream-side, take photograph vouchers. If gross morphological features will not be adequate, and the specimen is not potentially a member of a rare, threatened, or endangered species, collect 1-2 individuals in a container with 80% ethanol for laboratory identification.
10. Record non-biological observations and measurements at time of sampling. This will include weather conditions, flow and discharge observations, depth, substrate, % of pool, run and riffle, habitat quality, amount of time shocking (in seconds), water temperature, specific conductance, dissolved oxygen, turbidity, and pH; details regarding these observations and measurements are described in Section 3.6. Also, see VT DEC Fish Population Field Sheet 2021 (Appendix A, Figure A.4).



Table 6.7.A Minimum section lengths for sampling fish communities.

Mean Width	Minimum Reach Length	Number of Anodes
≤ 3.5 m	75 m	1
> 3.5 - 5 m	100 m	1
> 5 - 8* m	120 m	2
> 8 - 12* m	150 m	2
> 12**m	150 - 200 m	2 (bank shock)
*Consider using two shockers @ streams >6m wide		
**May include shocking both banks out to 6 meters.		

Table 6.7.B Electroshocker suggested voltage settings based on in-stream conductivity.

In-stream Conductivity	Electroshocker Volts setting
< 40	990
40-70	700
70-100	500
100-175	400
175-250	350
250-500	300
500-750	250
750-1500	200
> 1500	150
Suggested shocker settings based on conductivity (µS/cm)	

6.7.2 Lake Sampling

Seine Nets

Seines are generally used for strictly qualitative work. Their use is limited to waters of less than 1 m deep (3.2 ft), i.e. littoral shallows, where little or no bottom obstructions exist. Operation requires two people, one at each end, walking the net (which is parallel to shore) towards shore while maintaining net contact with the bottom and the float end of the net on the water surface. The netters move the net through the water up onto the shore trapping the fish in the net. Seine nets range in size from 4.6 to 15.2 m (15 – 50 ft) in length and are 1.2 m (4 ft) in height.

Minnow Traps

These devices are small wire mesh traps with holes at each end. When baited with dog food or bread, small fish find their way into the trap and cannot escape. Minnow traps are inexpensive and provide qualitative data only, these traps are generally set on the bottom at depths less than 2 m (6.5 ft). Minnow traps are also useful for surveying crayfish populations.

7. Wetlands

Wetlands are assessed by the Wetlands Bioassessment Program. The purpose of the Program is to assess wetland biological integrity and determine the ecological condition of Vermont's wetlands. Over the long term, it is expected that results from the program may be used for: improved permitting and regulatory decisions; providing significant information for mitigation and restoration projects; and identifying the effects of environmental and anthropogenic stressors on wetlands over time.

The Program utilizes three different survey types which is reflective of the EPA's Level 1, 2, and 3 approach to wetland monitoring. All three levels may be applied at any given site. Level 1 is a broad landscape-scale assessment performed as a desktop review using GIS, LiDAR and aerial imagery; Level 2 is a rapid field assessment at the wetland scale and are to be validated by and calibrated to Level 3 assessments; Level 3 is a site-intensive biological assessment using multi-metric indices. Chemical and physical data are collected as well. For full protocols, see the referenced documents below.

7.1 Level 2 VRAM Rapid Assessments

The Vermont Rapid Assessment Method (VRAM) for Wetlands is used for assessing wetland condition, function, and value. The 'Grand Total' score is based on a numeric scale ranging from 1 to 100 and provides the wetland's overall "quality score: The higher the score, the higher the quality of the wetland. An adapted VRAM protocol known as the "Indicators of Restoration Success" can also be used to monitor broad-scale wetland restoration success over time. Additionally, natural community mapping per the Vermont Natural Heritage Inventory is also performed for most wetland assessment sites. When appropriate and feasible, plant species lists for vegetation communities in wetlands are collected. For more information, please refer to "[Vermont Rapid Assessment Method for Wetlands v.2.2 User's Manual and Scoring Form](#)" last revised on 2/26/2019.



7.3 Level 3 Wetland Assessments

The Wetlands Program conducts detailed biological surveys which involves identifying plant species in a section of wetland for the Floristic Quality Assessment (FQA) analysis. The vegetation plots used are similar to Natural Heritage Inventory (NHI) methodology. Additionally, water chemistry parameters are collected, and the soils and hydrology are characterized for the wetland site. Other biocriteria have been developed based on plant species lists and VRAM data to infer additional characteristics of wetlands and their condition. For more information, please refer to the "Quality Assurance Project Plan (QAPP) for Biological Monitoring of Vermont's Wetlands: An Evaluation of the Chemical, Physical, and Biological Characteristics of Vermont Wetlands"; prepared by VT DEC; last revised 1/30/2019.

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Appendix

LOTIC BENTHOS FIELD SHEET (2021 edition)			
Stream Name _____ DEC Site ID _____ Date _____ Crew _____ Site/Access Description: _____		River Mile _____ New Site: Y N Latitude _____ Longitude _____ Site Type: WWTF USFS SENT PROB Other _____ Other Monitoring: Fish SGA-lite Temp Other _____	
Water Chemistry Sampler (lab parameters) _____ Flow Type: Baseflow Freshet Flow Level: High Medium Low Weather/Flow Comments (previous 2 days – 2 weeks) _____ Other Chemistry Comments _____ Lab Parameters: Chem Lab ID _____ Dup Lab ID (if applicable) _____ Time _____ Dup Time (if applicable) _____ Parameters collected (circle): Alk TP DP TN NOx NH3 Cl SO4 Turb DOC Earth Metals Total Metals Other _____ Field Parameters: Meter used _____ Water Temp (°C) _____ pH _____ Cond (uS) _____ Turbidity (NTU) _____ D.O.(%) _____ D.O. (mg/l) _____ Calibrated within 7 days (pH/Cond/Turb): Y N Calibrated on site (D.O.): Y N			
Macroinvertebrates Sampler _____ Method _____ Composites/Rep _____ Trophic Rating _____ (0=Oligotrophic, 5=Eutrophic) Comments _____ Low Gradient Habitat (if applicable): Poor Fair Good VG Exc Low Gradient Composites (sum = 4): Macrophytes _____ Woody snags _____ Root wads _____ Overhanging herbaceous _____ Overhanging branches _____		Stream Characteristics Habitat type: Riffle Low Gradient Other _____ Bankfull Width (m) _____ Wetted Width (m) _____ Velocity: Slow (<0.4 ft/sec) Medium (0.4-2 ft/sec) Fast (>2 ft/sec) Bank Stability: Poor Fair Good VG Exc Large Woody Debris (>4" diameter): # _____/100m	
Riparian Characteristics Riparian Width (facing upstream – 25m max): L _____(m) R _____(m) Canopy (estimate) _____% Overstory (>5m, should be </= 100% combined) : Softwood _____% Hardwood _____% Understory (<5m, can overlap and be >100%) : Woody/shrub _____% Herbaceous _____% Grass (lawn/pasture) _____%			
Substrate Characteristics Embeddedness (% estimate) _____ Silt Rating: _____ (0=none, 5=heavy plume) CPOM (leaf packs): _____ (0= none, 5=high) Periphyton Cover - Qualitative (See back for Substrate Periphyton Cover Form): Diatom _____% Filamentous Green _____% Blue Green _____% Moss _____% Green _____% Other _____% Calcareous Deposits _____% Iron Precipitate _____%			
General Habitat Observations and Comments (e.g. site sketch, aesthetics, land use, pollution, water clarity/color, etc)			

Figure A.1 Lotic Benthos Field Sheet

Site _____ Crew _____ Date _____

Pebble Count Field Form*(Complete "% Estimate" substrate only if no pebble count)*

Particle	Millimeters	% Estimate	Tally (100 minimum)	Total #
Clay	<.004			
Silt	.004 -0.6			
Sand	0.06 – 2.0			
Gravel	2.0 -16			
Coarse Gravel	16 – 64			
Cobble	64 – 256			
Boulder	>256			
Bedrock				
TOTALS:				

Periphyton Cover Observations

Moss Cover Index				
Category	0	1(<5%)	2(5-25%)	3(>25%)
Tally				

Macro-Algae Cover Index				
Category	0	1(<5%)	2(5-25%)	3(>25%)
Tally				

Micro-Algae Cover Index						
Category	0	1 (slimy)	2 (draw line)	3 (0.5-1mm)	4 (1-5mm)	5 (5-20mm)
Tally						

Other Percent Cover Index (e.g., Didymo, Riverweed)				
Category	0	1(<5%)	2(5-25%)	3(>25%)
Tally				

Other Thickness Cover Index (e.g., Iron precipitate, Calcareous deposits)						
Category	0	1 (slimy)	2 (draw line)	3 (0.5-1mm)	4 (1-5mm)	5 (5-20mm)
Tally						

Figure A.2 Pebble Count Field Sheet

VERMONT DEC FISH POPULATION FIELD SHEET (updated: 2021)

Location & River Mile _____	Personnel (circle ID) _____
Date _____ Time _____ Event ID _____	Section length (m) _____ Avg Width (m) _____
Biosite ID _____	Sample Type: Full section / Banks only (sampled width) _____
Comments:	Anode # _____ Volts: _____ PPS: _____ %Duty: _____
	Seconds: _____
	Run 1 Time _____ Run 2 Time _____ Run 3 Time _____

Species	Species Counts				Run ____ of ____	Anomaly Count	Total Count
AEL American Eel ABL American Brook Lamprey APD Allegheny Pearl Dace BAK Banded Killifish BCS Blackchin Shiner BND Blacknose Dace BNS Blacknose Shiner BRM Brassy Minnow BRS Bridle Shiner BSB Brook Stickleback BKT Brook Trout	BBH Brown Bullhead BRT Brown Trout BUR Burbot CAP Carp CHP Chain Pickerel CHD Channel Darter COS Common Shiner CRC Creek Chub CLM Cutlips Minnow	ESD Eastern Sand Darter EMS Emerald Shiner FAF Fallfish FTD Fintail Darter FHM Fathead Minnow FSD Finescale Dace GSH Golden Shiner LSPP Lamprey SA LMS Largemouth Bass	LOP Log Perch LND Longnose Dace LNS Longnose Sucker MSH Mimic Shiner MSC Mottled Sculpin MDM Mudminnow NOP Northern Pike NPD Northern Pearl Dace PHOX Phoxinus Hybrid PMS Pumpkinseed	RBT Rainbow Trout RBD Redbelly Dace RBS Redbreast Sunfish RES Redear Sunfish RFP Redfin Pickerel ROB Rock Bass RFS Rosyface Shiner RSD Rosyface Dace SSH Sand Shiner SEL Sea Lamprey SIM Silvery Minnow	SSC Slimy Sculpin SMB Smallmouth Bass SFS Spotfin Shiner STS Spottail Shiner STC Stonecat TED Tessellated Darter TRP Trout Perch WHS White Sucker YBH Yellow Bullhead YPE Yellow Perch		

Figure A.3 Fish Population Field Sheet

Vermont DEC Fish Section Habitat Evaluation Sheet

INSTREAM COVER: ~~Exc~~ VG G F P COVER TYPE: Boulders LWD Root Wads Overhang Veg Undercut Banks

NUMBER of LWD (>4") / 100m: _____ FISHABILITY: ~~Exc~~ VG G F P

BANK VEGETATION: _____ CANOPY (%): _____ BANK STABILITY: ~~Exc~~ VG G F P

DISCHARGE: H M L BOTTOM TYPE: hard soft mix GRADIENT: high med low flat

EMBEDDEDNESS: 0-5% Excel (5) 5-25% V Good (4) 25-50% Good (3) 50-75% Fair (2) >75% Poor (1)

Lab Chemistry Collected: Y / N

SAMPLE REACH % Pool: _____ % Run: _____ % Pool: _____

LENGTH AND WIDTH MEASUREMENTS	
LENGTHS	WIDTHS (6-10 meas)

Site Sketch

General observations:

Debris Obvious Pollution: Sludge, Sawdust, Paper Fiber, Sand, Silt, Sewage, Oily Sheen, Trash, Iron, Scum, None

Water Clarity: Clear, Slightly Turbid, Moderately Turbid, Very Turbid

Water Color: Clear, Green, Milky, Brown (Tannic) L M H, Gray, Metallic, Reddish

Comments:

Anomaly Comments:

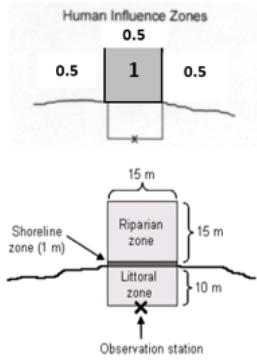
Conductivity	Volts
< 40	990
40-70	700
70-100	500
100-175	400
175-250	350
250-500	300
500-750	250
750-1500	200
> 1500	150
Suggested shocker settings based on conductivity (µS/cm)	

Mean Width	Minimum length to be sampled	Number of Anodes
≤ 3.5 m	75 m	1
> 3.5 - 5 m	100 m	1
> 5 - 8* m	120 m	2
> 8 - 12* m	150m	2-3
> 12**	150 - 200 m	2 (bank shock)
*Consider using two shockers @ streams >6m wide **May include shocking both banks out to 6 meters.		

Figure A.4 Fish Section Habitat Evaluation Sheet

2019 LAKE ASSESSMENT – PHAB SITES

Lake:			Site (A-J):			Date:			Crew:		
Site Type:	Point	Cove	Shore	Inlet	Other	Site Developed w/in 100m?	Yes	No	Lat:	Long:	
Habitat:	Rocky Littoral Macrophyte Muddy Littoral Sandy Littoral Other:								GPS projection:		
Snorkeler:			Macroinvertebrate Collector:			Buffer Meas By:			PHAB Measure By:		
NLA: RIPARIAN*											
Riparian Level	Vegetation Type		% Cover*		Type (circle one)	Riparian Level	Vegetation Type		% Cover*		*For NLA Riparian, percent cover for each vegetation type can range from 0-100%. If the cover of big trees is 100% and the cover of small trees is 70%, the total canopy cover is 170%.
Canopy (>5 m)	Big Trees (>0.3m DBH)				Decid Conif Mix	Ground Cover (<0.5 m hgt)	Woody Shrubs & Saplings				
	Small Trees (<0.3m DBH)				Decid Conif Mix		Non-woody Shrubs & Saplings				
Understory (0.5-5 m)	Woody Shrubs & Saplings				Decid Conif Mix		Standing H2O or Inundated Veg				
							Bare Ground				
LHA: IMMEDIATE SHORE TYPE**						NLA LAKESHORE DISTURBANCE & LAKES SCORECARD					
SHORE TYPE	% Cover**		Type (circle one)								
Tree Stratum (>5m)			Deciduous Coniferous Mixed								
High Shrub (1.5 – 5m)			Deciduous Coniferous Mixed								
Low Shrub (0.1-1.5m)			**% Cover must total 100% (e.g. TS=50, HS=20, LS=15, & GC=15)								
Ground Cover											
Site Photo Taken?	Yes	No									



HUMAN INFLUENCE	0, 0.5, 1	HUMAN INFLUENCE	0, 0.5, 1
Buildings		Lawn	
Park Facilities/ Man-made beach		Walls, Dikes, Revetments	
Commercial		Landfill/Trash	
Docks/Boats		Roads/Railroad	
Row Crops		Powerlines	
Pasture/Range/Hay Field		0 = NOT PRESENT 0.5 = PRESENT OUTSIDE PLOT 1 = PRESENT INSIDE PLOT	
Orchard			

Figure A.5 Lentic Physical Habitat Assessment Form

2019 LAKE ASSESSMENT – PHAB SITES

LAKE:		SITE (A-J):		DATE:	
MACROPHYTE FUNCTIONAL GROUPS		% Cover	LITTORAL HABITAT PARAMETERS	% Cover	
Submergent			% Sand		
Emergent			% Embeddedness		
Floating			Densiometer (1-17)		
DOMINANT 3 SPECIES		% Cover	INVASIVE SPECIES		
1.			LITTORAL PLOT SPECIES (AQUATIC)		% Cover
2.					
3.					
FISH COVER		% Cover			
Aquatic & Inundated Herbaceous Veg.					
Woody Debris/Snags >30 cm diameter			RIPARIAN PLOT SPECIES (TERRESTRIAL)		% Cover
Woody Brush/Debris <30 cm diameter					
Inundated Live Trees					
Overhanging Veg w/in 1m of surface					
Ledges or Sharp dropoff					
Boulders					
Human Structures – docks, buildings, etc					
Complexity of View Across Lake from Site		None	Low	Moderate	High
Photo Taken?		Yes	No		

Figure A.6 Lake Assessment Field Sheet

Littoral Habitat Assessment 2015 Datasheet

Lake Name		Date		SEDIMENT PLUGS	Indicate: YES / NO
Site # (note on map)		GPS Code		S @ 5M	
Site type: Point Cove Shore Inlet Other: _____		Terrestrial Sampler		M @5M	
Habitat: Rocky Littoral Macrophyte Muddy Littoral Sandy Littoral Other: _____		Plant Snorkeler		D @5M	
Site Development (structure w/in 100M of shore?): Yes or No		Woody Debris Snorkeler		DISTANCE FROM SHORE TO 2M depth (m) Pole / Rangefinder (circle one)	
If Yes: Recent or Established?		Latitude:		2A (start) to shore:	
		Longitude:		2C (start) to shore:	
				DENSIOMETER @ transect midpoint (0-17)	
				1M from shore:	
				5M from shore:	

+ Riparian Vegetation % Cover (high water mark to 10M inland)

Immediate Shore Type	% Cover	Circle One			To 10 M inland, dominant ground cover type (duff, lawn, etc.)		
Tree Stratum (>5 m hgt)		Deciduous	Coniferous	Mixed			
High Shrub (1.5-5 m hgt)		Deciduous	Coniferous	Mixed			
Low Shrub (0.1-1.5 m hgt)		# CWD (>10 cm):		# Odonate Exuviae:	# Tubes:		
Ground cover							
PLOT PARAMETERS		S1 Plot % Cover	S2 Plot % Cover	M1 Plot % Cover	M2 Plot % Cover	D1 Plot % Cover	D2 Plot % Cover
Woody Debris	Fine (<4cm)						
	Medium (4-10cm)						
Deciduous Leaf Litter							
% Embeddedness							
Fish (mark if observed)							
Mussels							
Crayfish							
Snails							
Macroinvertebrates sampled? Yes No				Habitat Type: Sandy Rocky Muddy Macrophyte			
Method: 10cm-Deep-Core Rock-Washing							

Figure A.7 Littoral Habitat Assessment Data Sheet

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