

# North Carolina Aquatic Data Hub Methods Manual





## **Prepared by the North Carolina Aquatic Data Hub**

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Contributors to the NCADH include:

Carolina Wetlands Association

Environmental Quality Institute

Haw River Assembly

Morehead Planetarium and Science Center

MountainTrue

NatureServe

New River Conservancy

North Carolina Department of Environmental Quality - Division of Water Resources

North Carolina Museum of Natural Sciences

North Carolina Natural Heritage Program

North Carolina Watershed Stewardship Network

River Guardian Foundation

River Network

UNC Institute for the Environment

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# Table of Contents

<b>Acknowledgements</b>	<b>5</b>
<b>About the NC Aquatic Data Hub</b>	<b>6</b>
<b>Objectives and goals</b>	<b>6</b>
<b>How to use this manual</b>	<b>7</b>
<b>Safety considerations</b>	<b>8</b>
<b>Selecting a site</b>	<b>10</b>
<b>Sampling at different location types</b>	<b>11</b>
<b>Data tiers</b>	<b>12</b>
<b>Tiered data quality objectives</b>	<b>13</b>
<b>Holding times</b>	<b>16</b>
<b>List of terms</b>	<b>17</b>
<b>NCADH Data Sheets</b>	<b>20</b>
<b>Habitat assessment</b>	<b>21</b>
<b>BAU Habitat assessment</b>	<b>25</b>
<b>Chemical/Physical Protocols</b>	<b>29</b>
Collecting a water sample	29
Grab samples for lab analysis	30
pH: Colorimetric Kit	34
pH: Test Strips	35
pH: Pen Meter	37
pH: Probe	39
Water/air temperature: Handheld Thermometer	40
Water/air temperature: Probe	41
Fecal coliform: Coliscan Easygel	42
Fecal coliform: Grab samples for lab analysis	45
Dissolved Oxygen: CHEMetrics ampoules	47

Dissolved Oxygen: Pen meter	48
Dissolved Oxygen:probe	49
Water clarity: turbidity tube	52
Conductivity: Pen meter	53
Salinity: Pen meter	54
Chlorophyll A:                      Grab samples for lab analysis	55
<b>SOS Benthic macroinvertebrate assessment</b>	<b>56</b>
SOS Macroinvertebrate Tally Sheet	64
<b>SMIE Benthic macroinvertebrate assessment</b>	<b>66</b>
<b>BAB Benthic macroinvertebrate assessment</b>	<b>71</b>
<b>Appendix 1. Temperature Conversion Table</b>	<b>72</b>
<b>Appendix 2. Equipment Information</b>	<b>73</b>
<b>Appendix 3. Example Equipment Resolution and Accuracy</b>	<b>74</b>
<b>Appendix 4. Turbidity tube construction and CM to NTU Table</b>	<b>75</b>

## Acknowledgements

Contents of this manual were adapted from the following sources:

Chesapeake Monitoring Cooperative, 2016, Non-Tidal Methods Manual

Environmental Quality Institute, 2018, Stream Monitoring Information Exchange: Ecological Stream Monitoring Protocol for Western North Carolina Wadeable Streams and Rivers

Georgia Adopt-a-Stream, Habitat assessment

Indiana Department of Environmental Management, 2017, Volunteer Stream Monitoring Training Manual

Issac Walton League Save Our Streams, benthic macroinvertebrates

New River Conservancy, 2015, New River Water Watchers Volunteer Water Quality Monitoring Program Manual

North Carolina Department of Environmental Quality Division of Water Resources, 2012, Monitoring Coalition Program Field Monitoring Guidance

North Carolina Department of Environmental Quality Division of Water Resources, 2013, Intensive Survey Branch Standard Operating Procedures Manual: Physical and Chemical Monitoring

US Environmental Protection Agency, 1997, Volunteer Stream Monitoring: A Methods Manual

Virginia Citizen Water Quality Monitoring Program, 2007, Virginia Citizen Water Quality Monitoring Program Methods Manual

# About the NC Aquatic Data Hub

The North Carolina Aquatic Data Hub is a new initiative for connecting aquatic monitoring efforts across the state in order to better understand the condition of North Carolina's waters and to maintain and improve them. NCADH provides the resources and training for new groups and existing organizations to contribute to and access a statewide network of aquatic data.

The North Carolina Aquatic Data Hub has been made possible by a \$160,000 two-year grant awarded by the Z. Smith Reynolds Foundation to support a statewide citizen science water quality monitoring project to New River Conservancy on behalf of multiple non-profits and agencies across the state. In April of 2018, NRC successfully applied for a National Fish and Wildlife Foundation NC and VA River and Waters Program Grant.

Partners of this initiative include River Network, NC Watershed Stewardship Network, Carolina Wetlands Association, NC Museum of Natural Sciences, Morehead Planetarium & Science Center, NC Division of Water Resources, Water Resources Research Institute of the UNC System, UNC Institute of the Environment, River Network, Environmental Quality Institute, and NC Natural Heritage Program.

The need for a citizen science water quality monitoring program was highlighted at the Water Education Summit in Asheville in September 2014, during a panel discussion regarding successful volunteer biomonitoring programs and the opportunities for a statewide approach for monitoring. Panel participants were extremely receptive to the idea of a statewide program for NC. They recognized 1) monitoring and 2) database management as important tools that need to be better developed if we are to more effectively address water quality and biodiversity concerns in our region's watersheds.

Recognizing the value of monitoring programs as effective tools towards reaching conservation action, the development of a statewide monitoring program is a prime opportunity to improve (or at least maintain) NC waters, especially in an era of decreased government funding for monitoring.

## Objectives and goals

- Improve accessibility of citizen data to the public and to state and federal agencies
- Create greater citizen involvement in aquatic monitoring
- Increase awareness of the health of North Carolina's waters
- Enhance watershed management efforts

# How to use this manual

The Table of Contents is separated by sections. Use these sections as a guide during each monitoring event. Your monitoring efforts will be based on three parameters: habitat assessment, benthic macroinvertebrates, and chemical/ physical. The manual is grouped by parameter, then subgrouped by tier. If you are unsure what tier method you are using, see section (# or letter), “Data Tiers,” or contact your local NCADH affiliate.

In section (# or letter), “Method Protocols,” you will find detailed instructions on each element of your monitoring effort. Icons for each monitoring parameter can be found on the top left corner of each protocol page. Within each protocol, you will find instructions for Tiers 1 - 3.

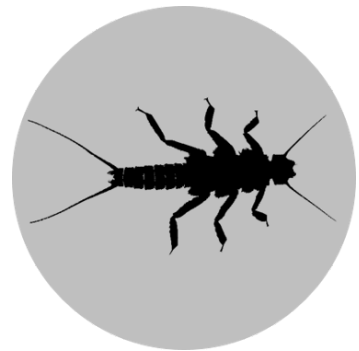
Most measurements can be performed using more than one method and in multiple tiers. To help you keep track of which tier a particular method belongs to, each method listed will include icons to indicate tier and sampling type, as shown below. Red diamonds indicate Tier 1 methods, blue rectangles indicate Tier 2, and green ovals indicate Tier 3 methods. Additionally, headings in the section describing a particular method are in the color of the icon. For example, headings in a section for a Tier 2 method would be written in blue text.



**Habitat assessment**



**Chemical/physical**



**Benthic macroinvertebrate**





# Safety considerations

Streams and riparian zones can be dangerous places if the proper precautions are not taken to prevent mishaps. Safety takes priority over data or sample collection, even if it is inconvenient or you have to abandon gear. Upon arriving at the stream, take an extra minute or two to identify any potential threats. If you do locate problems, consider changing the sampling location or cancel/reschedule sampling. Bring the phone number of the program coordinator into the field in case guidance is needed. Record safety concerns on monitoring data sheets.

Check weather reports before going into the field. Sampling in the rain is fine as long as volunteers are comfortable, but be aware of threats from lightning and flash flooding. If it's too cold, cancel and reschedule. You can also take breaks in a warm car. Signs of hypothermia include blue lips, trouble thinking clearly, and bouts of uncontrolled shaking.

Wading should be done with caution, as a slip midstream can make for a miserable wet sampling experience and could lead to further complications. Avoid fast currents above the knees and wear closed-toed shoes/boots/waders that protect your feet from jagged rocks and broken glass. Foot entrapment by debris, rocks, or muck can be a severe hazard in deeper water. Be careful of going too deep with chest waders since they can also be a drowning hazard if they fill with water. Walking sticks can be used to provide additional stability and test the substrate if turbid water obscures the stream bottom.

In addition to wild animals and plants, humans and aggressive dogs can pose safety concerns. Efforts should be made to ensure monitoring sites are accessed via public right-of-way (generally a public park or 50 feet to either side of state-maintained bridges) or with permission from a landowner. If sampling from a bridge, be aware of moving vehicles. Sample with a partner, especially if the site is not located in a populated location. Do not discount the safety concerns of any member of a sampling team. Let someone know where you are going and when you expect to return.

If sampling with chemicals, familiarize yourself with the proper uses and dangers associated with each. Label all chemicals clearly. Bring proper safety equipment as needed, such as protective gloves, eye protection, and a container for sharps or waste. Clean up any spills and dispose of waste in a safe manner. Keep chemicals away from children and pets.

Personal safety items include things individuals should bring to protect their own health. This can be a general first aid kit or medication for allergies or blood sugar disorders. Water is a requirement to avoid dehydration while in the field. Sunscreen, sunglasses, a hat, and insect repellent should be brought at the discretion of the individual. Appropriate clothing is also a must in river sampling, so consider bringing extra socks, multiple layers of clothing, warm clothing (avoid cotton as it will not keep you warm if it becomes wet), and a rain jacket. Hand

sanitizer should be brought for use before handling food or tobacco products after exposure to stream water.

**Potential dangers may include:**

Plants

- Poisonous plants (e.g. poison ivy, poison oak, poison sumac)
- Stinging plants (e.g. stinging nettle)
- Thorny plants (e.g. blackberry vines, “briars”)

Insects

- Stinging insects (e.g. bees, wasps, hornets)
- Biting insects (e.g. fire ants, mosquitoes, ticks, chiggers, biting flies)
- Spiders

Animals

- Snakes (e.g. cottonmouths in the lower Piedmont and Coastal Plain)
- Aggressive dogs
- Livestock on farmland
- Humans
- Large wild animals (e.g. alligators, bears)
- Any animal behaving in an unusual manner

Stream

- Fast currents
- Flash flooding
- Rocks/boulders
- Slippery substrate and steep streambanks
- Unconsolidated mud/muck/silt
- Deep pools

Other environmental concerns

- Lightning
- Flash flooding
- Cold/heat
- Sun
- Bacteria/pathogens from stream water
- Trash (e.g. broken glass, metal)

# Selecting a site

The first step in choosing a monitoring site is to ask what is to be achieved by monitoring. If trying to measure the impact of suspected point-source pollution, locate sites upstream and downstream of the suspected source. If measuring nonpoint-source pollution, the base of the tributaries within the watershed or subwatershed may be the most appropriate locations. If community outreach is a priority, public parks or schools may make the monitoring more accessible. Human health concerns may spur monitoring at recreation hot-spots (fishing, swimming, boating, etc.). Collaboration with regional stakeholders, such as local governments, nonprofits, or community groups, may help to identify sites where monitoring data can yield the biggest impact. NC DEQ monitoring sites offer opportunities to compare volunteer data to state-generated data. Sites located in pristine, forested, or minimally impacted areas are important for comparison with sites located in areas of concern. Sites may also be established downstream of existing or planned stream restoration and mitigation projects to capture short or long-term performance.

Many monitoring projects use fixed sampling locations to focus on changes over time, season, and streamflow. Rapid-response sampling is employed for short-term monitoring to document specific threats, such as construction projects, stormwater runoff, chemical spills, etc.

All sampling should take place on public property, such as a park, bridge, or pier/dock, unless the landowner's permission is documented. No trespassing is allowed for either sampling or parking. Room for parking in a lot or on the shoulder of a road should be available. Volunteers need reasonable access to the stream for sampling, with minimal environmental hazards.

There are additional considerations for biological monitoring sites, such as for benthic macroinvertebrate sampling. For the wadeable stream protocols, the sites should have a stream width of at least 6 feet (2 meters) across, preferably with riffle habitat present. There should be room on the banks to set up a processing and/or identification station with a folding table. With a larger sampling team, there should be parking space for more than one car.

Once a site is selected, the following information should be documented:

- Location name and description
- Latitude/longitude
- Representative Photographs
- Detailed driving and stream access instructions
- Initial habitat assessment

# Sampling at different location types

Water sampling techniques will depend on the depth and width of the stream and whether it is wadeable or non-wadeable. The goal is to collect a representative sample from flowing water in the main channel. Wadeable streams may be accessed with waterproof boots and waders, or even from the shoreline for very small waterways. A bridge, boat, dock, or pier can provide access for non-wadeable streams. A telescoping pole sampler can help sample streams that are too deep for wading but not very wide.

When wading into a stream try to minimize sediment disturbance, especially in low-flow conditions. Sample the water upstream of where you are standing to avoid collecting disturbed sediment. Collect from the middle of water column with the bottle opening facing upstream.

Sample upstream also if collecting from a dock, pier, or boat. From a bridge, collect stream water with either a bucket or a weighted basket sampler containing your bottles, lowered with a rope. If using a bucket, beware of sediment settling to the bottom before you are able to pour the water into sample bottles or directly analyze it.

Prior to water sampling, obtain clean bottles or wash them appropriately depending on the analysis. Bottles used for bacteria analysis must be sterilized prior to sample collection. Label sample bottles with permanent markers or pencil on labeling tape. Avoid touching the inside of sample containers. If using chemical preservative, you may use a clean transfer bottle to fill the sample bottle without losing preservative. Leave headspace when filling the bottles with water unless the method says otherwise (e.g. DO, BOD).

Benthic macroinvertebrate sampling protocols described in this manual are only conducted in wadeable streams. Ideally, kick nets and leaf packs are used for sampling in riffle habitats. D-frame nets or sweep nets are used in lower flow streams with no riffles present. Visual assessments are important for recording invertebrates present in other habitats such as bank roots, pools, organic debris, logs, and rocks.

More detailed descriptions of sampling techniques are available in the following method protocols.

# Data tiers

Method Tier	Data uses	QA/QC Requirements
<i>Tier 3</i>	Assessment	Data collection follows protocols used by NCDEQ to list and delist waterbodies. DWR Approved QAPP needed
<i>Tier 2</i>	Identify waters for follow up monitoring	QAPP required and followed
<i>Tier 1</i>	Education, baseline data, "red flags"	QAPP is not required

The tier level is determined by both the resolution and accuracy of the equipment used and the Quality Assurance Project Plan (QAPP).

For example an expensive highly accurate meter used without an accepted QAPP would only qualify as Tier 1 data.

# Tiered data quality objectives

The following table lists acceptable equipment types and precision ranges for each monitoring parameter and data tier.

Please note that the following is not an exhaustive list of equipment types that may be used, and that higher quality methods may be deployed at lower tiers if methods and quality are not documented with a Quality Assurance Project Plan. A table of example equipment and vendors is listed in Appendix 3.

	Tier 1 Protocols		Tier 2 Protocols		Tier 3 Protocols	
Parameter	Acceptable Equipment Types	Acceptable Precision Range	Acceptable Equipment Types	Acceptable Precision Range	Acceptable Equipment Types	Acceptable Precision Range
Site description	NCADH Rapid Visual Form	NA	NCADH Rapid Visual Form	NA	NC DWR Stream Habitat Assessment	NA
Stream habitat assessment	Adopt-A-Stream protocol (GA/SC or VA version)	NA	Adopt-A-Stream protocol (GA/SC or VA version)	NA	NC DWR Stream Habitat Assessment	NA
Temperature	Thermometer Meter	± 1° C	Thermometer Meter	± 0.5° C	Thermometer Meter	<a href="#">Link</a>
pH	Colorimetric strips Colorimetric test kits Meter	± 1 pH unit	Colorimetric strips Colorimetric test kits Meter Grab/lab sample	0.1 pH unit	Meter Grab/lab sample	<a href="#">Link</a>

<b>Dissolved Oxygen</b>	Colorimetric test kits	$\pm 2$ mg/L	Colorimetric test kits	$\pm 0.5$ mg/L	Meter	<a href="#">Link</a>
	Titration test		Titration test			
	Meter		Meter			
<b>Conductivity/ Specific Conductance</b>	Meter	$\pm 10$ $\mu$ S/cm	Meter	$\pm 1$ $\mu$ S/cm <sup>2</sup>	Meter	<a href="#">Link</a>
<b>Alkalinity</b>	Streamside test kit	$\pm 10$ mg/L	Streamside test kit	$\pm 5$ mg/L	Grab/lab sample	<a href="#">Link</a>
			Grab/lab sample			
<b>Coliform bacteria / E. coli</b>	Petri dish test kits	NA	Petri dish test kit Colilert - IDEXX Lab filtration method	NA	Colilert - IDEXX Lab filtration methods	<a href="#">Link</a>
<b>Chlorides/Salinity/ TDS</b>	Streamside test kit	$\pm 10$ mg/L	Streamside test kit	$\pm 1$ mg/L	Meters Grab/lab sample	<a href="#">Link</a>
	Meter		Meter			
<b>ChIA</b>	Meter	$\pm 0.1$ mg/L	Meter	$\pm 0.1$ mg/L	Meter Grab/lab sample	<a href="#">Link</a>
	Grab/Lab sample		Grab/Lab sample			
<b>Turbidity/ Water Clarity</b>	Turbidity tube Secchi Depth	$\pm 1$ cm	Turbidity tube	$\pm 1$ cm  $\pm 1$ (NTU, etc.)	Meter	<a href="#">Link</a>
			Secchi Depth			
			Meter			

<b>Nitrates</b>	Colorimetric strips	$\pm 1$ mg/L	Test kit	$\pm 0.1$ mg/L	Meter	Grab/lab sample	<a href="#">Link</a>
	Colorimetric test kits		Meter				
<b>Phosphates</b>	Colorimetric strips	$\pm 1$ mg/L	Grab/lab sample	$\pm 0.01$ mg/L	Grab/lab sample	Grab/lab sample	<a href="#">Link</a>
	Colorimetric test kits						
<b>Metals</b>	NA	NA	Grab/lab sample	$\pm 0.01$ mg/L	Grab/lab sample	Grab/lab sample	<a href="#">Link</a>



# Holding times

<b>Parameter</b>	<b>Maximum sample holding time</b>
Bacteria	24 hours
Clarity	Analyze on site only
Conductivity	28 days
Dissolved oxygen	Analyze on site only
Nitrate	48 hours
pH	24 hours
Temperature	Analyze on site only
Total dissolved solids	28 days
Turbidity	24 hours

# List of terms

**BMP** - Best Management Practice. Devices or systems that control construction and post-construction stormwater

**CWA**- Clean Water Act

**DWR**- Division of Water Resources (<https://deq.nc.gov/about/divisions/water-resources>)

**EPA** - Environmental Protection Agency

**HUC** - Hydrologic Unit Code

**NC DEQ**- North Carolina Department of Environmental Quality (<https://deq.nc.gov/>)

**NPDES** - National Pollution Discharge Elimination System. These permits regulate flow and constituents of a waste stream from point source dischargers into waters of the US. (Overview: <https://deq.nc.gov/about/divisions/water-resources/water-resources-permit-guidance/npdes-industrial-stormwater/history-water-quality-overview>) (Permit assistance: <https://deq.nc.gov/about/divisions/water-resources/water-resources-permit-guidance/npdes-industrial-stormwater/history-water-quality-overview>)

**NOV** - Notice of Violation. State and local permitting programs will issue NOV's for permit violations prior to a fine or other regulatory action in some cases.

**NTU** - Nephelometric Turbidity Unit. Measurement of Turbidity collected by quantifying amount of scattered light penetrating a water sample

**QA/QC**- Quality Assurance/ Quality Control

**QAPP** - Quality Assurance Project Plan

**SOP** - Standard Operating Procedures

**S&EC** - Sediment and Erosion Control. Most counties and jurisdictions have locally delegated S&EC programs, though some default to state regional offices.

**SPCA** - Sediment Pollution Control Act

**Surface Water Classifications** - NC DEQ applies classifications to streams, rivers, and lakes which define the best uses to be protected within these waters (such as fishing or water supply watersheds) and carry with them an associated set of water quality standards to protect those uses. (More information:

<https://deq.nc.gov/about/divisions/water-resources/planning/classification-standards/classifications>) (Interactive Online Map:

<https://ncdenr.maps.arcgis.com/apps/webappviewer/index.html?id=6e125ad7628f494694e259c80dd64265>)

**SW** - Stormwater

**TMDL** - Total Maximum Daily Load. When a waterbody is listed as 'Impaired' by the NC DEQ, the State is required by law to create a strategy to mitigate the impairment through a TMDL. (More information:

<https://deq.nc.gov/about/divisions/water-resources/planning/modeling-assessment/tmdls>)

(Interactive Online Map:

<https://ncdenr.maps.arcgis.com/apps/webappviewer/index.html?id=bc125c8b5ccf4110b538db1188731690>)

**WS** - Watershed. All area that drains into a specific waterbody

**WQS** - Water Quality Standards. NC DEQ sets water quality standards for pollutants and other parameters to protect a waterbodies designated use or classification.

(<https://deq.nc.gov/about/divisions/water-resources/planning/classification-standards/surface-water-standards#WQSTables>)

**WWTP**- Wastewater treatment plant. Some jurisdictions separate municipal from industrial wastewater, but not all. Water is treated and returned to a body of water

**USGS** - US Geological Survey

**303(d) List** - This list is comprised of category 5 (Impaired) waters of the water quality assessment. Category 5 waters do not meet the state water quality standards and are in need of a TMDL. Category 4 waters also do not meet state water quality standards but either have a TMDL or alternative to a TMDL in place.

More information:

<https://deq.nc.gov/about/divisions/water-resources/planning/classification-standards/303d/303d-files>

Interactive Online Map:

<https://ncdenr.maps.arcgis.com/apps/webappviewer/index.html?id=dcb44280272e4ac49d9a86b999939fec>

# NCADH Data Sheets

Different datasheets are provided to complete different types of stream assessments. This information should help you determine the correct datasheets to use for your selected protocols.

## Site descriptions

NCADH Rapid Visual Form

This form should be completed on every visit, no matter which additional survey methods you are choosing to conduct. This form helps us capture the necessary metadata (data that documents how things were accomplished) that is needed to ultimately make your data useful to others.

## Stream habitat assessments

[NCADH Adopt-A-Stream Visual Habitat Form](#)

Use this form to conduct Tier 1 & 2 stream habitat assessments. We recommend conducting these assessments on an annual basis.

[NCDWR Habitat Assessment Field Data Sheet](#)

This form can be used to conduct habitat assessments as part of Tier 1, 2, & 3 monitoring programs that want to collect state comparable data. Two different versions are deployed, one for Mountain and Piedmont regions and another for Coastal Plain streams.

[Virginia Save Our Streams Habitat Assessment Form](#)

Those monitors wishing to collect data that matches Virginia SOS protocols for river drainages extending into the state of Virginia may use the VASOS habitat assessment data sheet.

## Macroinvertebrate assessments

[Save Our Streams Macroinvertebrate Tally Data Sheet](#)

This datasheet is used to tally the types and numbers of macroinvertebrates observed during sampling and calculate a final index score of aquatic life health.

[Environmental Quality Institute SMIE Identification Data Sheet](#)

This datasheet is used to tally identifications of collected macroinvertebrates using the Stream Monitoring Information Exchange Protocol.

## Water quality / chemistry / bacteria

NCADH provides datasheets to track in-situ water quality collected at the streamside and forms for tracking grab samples collected in the field. A model Chain-of-custody Form is also provided.

## NORTH CAROLINA AQUATIC DATA HUB – STREAM VISUAL SURVEY

LOCATION INFORMATION:		TODAY'S SAMPLING EVENT:	
Stream Name:		Group/Org Name:	
Station ID:		Group/Org ID:	
NCADH Site ID:		Date:	
Latitude:		Start Time:	
Longitude:		End Time:	
GPS Device:		TODAY'S PERSONNEL:	
Location/Road:		TODAY'S SAMPLING TYPES:	
County:			Visual survey (below)
Watershed:			Photographs
Description/Comments:			Habitat assessment
			Water quality (field)
			Grab samples
			Macroinvertebrates
			Fish survey
			Algae survey/sample
			Bacteria sample
			Other: _____

### RAPID VISUAL STREAM SURVEY

WEATHER CONDITIONS: (LAST 24 HOURS)		WATER ODORS: (CHECK ANY THAT APPLY)		WATER FLOW STATUS: (CHOOSE ONE)		WATER SURFACE: (CHECK ANY THAT APPLY)	
Clear/ Sunny		None/natural		Dry – no water		Clear	
Partly cloudy		Sewage / Rotten egg		Standing pools		Oily sheen	
Overcast		Dead fish / animal		Low – Channel <25% full		- continuous film	
Intermittent rain		Chemical / chlorine		Normal – 25% - 75% full		- breaks easily	
Steady rain		Gasoline		High – Channel >75% full		Light foam	
Heavy rain		Other: _____		Flood – Water over banks		Foam chunks > 3 in.	

AQUATIC LIFE OBSERVED: (CHECK ANY THAT APPLY)		WATER COLOR: (CHECK ANY THAT APPLY)		OBSERVED POLLUTION /IMPACTS: (CHECK ANY THAT APPLY)		WATER CLARITY: (CHECK ANY THAT APPLY)	
Algae		Clear / No color		Vertical exposed banks		Clear / Transparent	
Aquatic plants (SAV)		Brown/Muddy		Sediment deposits		Cloudy / Somewhat turbid	
Fish		Black/Tannic		Evidence of floods			
Birds		Green		Trash		Opaque / Turbid	
Reptiles		Milky/white		Cows in stream		Other: _____	
Amphibians		Orange		Fish-passage barrier			
Mammals				Pipe or outfall			
Other: _____		Other: _____		Other: _____			



## Habitat assessment



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### ABOUT THIS HABITAT SURVEY

The Adopt-A-Stream habitat survey, protocol, and instructions have been adopted from the Georgia Adopt-A-Stream program. An excellent in-depth manual for completing this form can be found on the GA Adopt-A-Stream website:

[https://adoptastream.georgia.gov/sites/adoptastream.georgia.gov/files/related\\_files/document/Visual.pdf](https://adoptastream.georgia.gov/sites/adoptastream.georgia.gov/files/related_files/document/Visual.pdf)

### INTRODUCTION

Stream habitat includes the physical and chemical conditions of this ecosystem and plays a large role in the aquatic life you will find. By conducting this survey, you will be able to qualitatively document the condition of instream habitat and the riparian zone.

By conducting this on an annual basis, changes over time can be observed, paired with good snapshots upstream, downstream and into the riparian zone on both sides of the channel. The survey rates parameters including channel bottom materials, sinuosity, bank stability, streamside vegetation and many more. It is intended for wadeable streams only, and it is recommended that you read this guide first before completing the survey.

### USING THE SURVEY

Before you begin conducting the survey, there are some important concepts and ideas to keep in mind:

**Stream Reach:** At your adopted site, determine a section of stream you will walk and survey. The reach is defined as twelve (12) times the average width of the stream or at least 100 meters.

Figure out the most upstream and downstream points, and perhaps place a permanent marker so you remember this area for future surveys. Be sure to walk the entire reach, getting in the stream and riparian zone when completing the survey to achieve a better idea of the overall condition.

**Reference Stream:** This survey works best if you have identified a local reference stream for comparison. This is a stream that has had minimal disturbance from human interactions, or is as healthy a stream you

can find in your area and which can serve as a benchmark for the survey and other streams you evaluate.

**Total Score:** After you have evaluated and scored each parameter, sum up the total points to determine your final score and rating for your stream.

**Rocky Bottom vs. Muddy Bottom Streams:** Coastal streams are classified as muddy bottom, while Piedmont and Mountain streams are rocky bottom. Evaluate only those parameters that are appropriate for that type (i.e. #2 refers to only rocky bottom streams, #7 refers to only muddy bottom streams).

**Photo Points:** Take four (4) images of your site, 1 upstream, 1 downstream, 1 looking at the left bank/riparian zone, and 1 looking at the right bank/riparian zone. Take a set of these each time you conduct the habitat survey.

### SCORING THE PARAMETERS

Each habitat parameter is rated with a value from 0 to 10, and in some cases (Numbers 8, 9 and 10) you will be asked to evaluate each bank separately, scoring each from 0 to 5.

Using the data form, record the score that best fits the observations you have made based on the narratives, drawing, images and description provided in the following page of this guide. See the example form at the end of this document to assist you in completing the survey.

### HOW DO I INTERPRET MY TOTAL SCORE?

This can change through the seasons and in varying weather and climatic patterns. It is good to have baseline data through the seasons and over time compare the total. It is also good to look at each parameter individually over time and perhaps see if there are interventions such as restoration initiatives that can be utilized. Additionally, if the survey is completed in conjunction with the macroinvertebrate survey, this should give insight into the results of the water quality index score in regards to overall habitat condition and habitat availability at your site.

### STREAM HABITAT SURVEY GUIDE

You will find a more in-depth guide to scoring each parameter, including what to look for, why is it important, how to score the parameter and a definition of terms on [NCAquaticDatahub.org](http://NCAquaticDatahub.org).









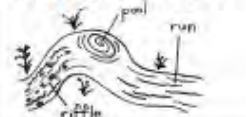








# NC AQUATIC DATA HUB: Stream Habitat Survey

(Also fill out the Basic Visual Form when completing this survey)

Type of Stream:  Rocky bottom  
 Muddy bottom

Stream habitat is evaluated looking both upstream and downstream, and includes: channel bottom materials, streamside vegetation, slope, and other channel characteristics. You may choose a value between 0-10 for each parameter. Note #s 8-10 ask you to evaluate each bank separately.

All measurements should be taken during baseflow conditions. Stream reach is defined as 12 times stream width, bankfull to bankfull.

Stream name:		Date:		Group/Org:	
Station ID:		Time:		Your name:	
<b>Habitat Parameter</b>	Excellent ----- -Poor				
<b>1. Epifaunal Substrate</b>	Abundant stable habitat cover for colonization by macroinvertebrates and fish: submerged roots, woody and vegetative debris, cobbles, leaf packs and undercut banks.	Adequate stable habitat cover for colonization by macroinvertebrates and fish: submerged roots, woody and vegetative debris, cobbles, leaf packs and undercut banks.	Little or no stable habitat cover available for colonization by macroinvertebrates and fish: submerged roots, woody and vegetative debris, cobbles, leaf packs and undercut banks; habitat may move during high flows.	What did you see?	
<b>What types of submerged materials are on the channel bottom?</b>					
	10 9 8 7	6 5 4 3	2 1 0	Score	
<b>2. Embeddedness</b>	Gravel and cobble are slightly embedded in riffle area.	Gravel and cobble are partially embedded in riffle area.	Gravel and cobble are completely embedded in riffle area.	What did you see?	
<b>* ROCKY BOTTOM streams only Are fine sediments being deposited in riffle/run area?</b>					
	10 9 8 7	6 5 4 3	2 1 0	Score	
<b>3. Riffle/Run/Pool</b>	Yes, all three (3) habitat types (riffle, run, pool) are present and frequent.	Two (2) habitat types are present.	Only one (1) habitat type present and dominant.	What did you see?	
<b>Is a diversity of instream habitats available: riffle, runs and pools?</b>					
	10 9 8 7	6 5 4 3	2 1 0	Score	
<b>4. Sediment Deposition</b>	Point bars and islands stable and of small size and frequency with some vegetation. Composed mostly of gravel and cobble.	Point bars and islands less stable and of moderate size and frequency with some sparse vegetation. Composed mostly of some gravel and finer sediment.	Point bars and islands unstable and of a large size with little or no vegetation. Composed almost entirely of fine sediment.	What did you see?	
<b>Are point bars and islands present?</b>					
	10 9 8 7	6 5 4 3	2 1 0	Score	
<b>5. Channel Flow Status</b>	Water reaches base of both lower banks; little substrate exposed.	Some substrate is exposed and water partially fills channel.	Most substrate is exposed and very little water in channel.	What did you see?	
<b>How much water is in the stream channel?</b>					
	10 9 8 7	6 5 4 3	2 1 0	Score	

Total first side \_\_\_\_\_

Habitat Parameter	Excellent -----Poor											
<b>6. Channel Alteration</b>												
<b>Is the stream channel altered by humans?</b>	No evidence of channelization (straightening) or alterations such as dredging, agriculture, concrete banks or construction activities.	Some evidence of channelization (straightening) and/or alterations such as dredging, agriculture, concrete banks or construction activities.			Most of stream reach channelized and/or many alterations present such as dredging, agriculture, concrete banks or construction activities.				What did you see?			
	10 9 8 7	6 5 4 3	2 1 0	Score								
<b>7. Channel Sinuosity</b>												
<b>Does the channel have lots of curves and bends?</b>	Yes, bends in the channel are frequent.	There are more bends than straight sections.			There are more straight sections than sections with bends or channel is entirely straight.				What did you see?			
* For MUDDY BOTTOM streams only												
	10 9 8 7	6 5 4 3	2 1 0	Score								
<b>8. Bank Stability</b>												
<b>How stable are the streambanks?</b>	Bank stable; erosion, scouring, undercutting or bank failure absent or minimal. Vegetation overhanging the stream is abundant.	Bank moderately stable; evidence of small areas of erosion, undercutting and scouring, or bank failure present. Moderate amounts of overhanging vegetation present.			Bank unstable; many eroded and scoured areas with undercutting; bank failure present; steep banks. Little overhanging vegetation present.				What did you see?			
Determine right/left bank by facing downstream												
Left bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	Score (Add both banks)
Right bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	
<b>9. Vegetative Protection</b>												
<b>Are streambanks covered &amp; shaded by a variety of vegetation?</b>	Most streambank surfaces covered and shaded by a large variety of vegetation (trees, shrubs, flowering plants and grasses).	Some streambank surfaces covered and shaded by some variety of vegetation (trees, shrubs, flowering plants and grasses).			Few streambank surfaces covered and shaded by vegetation. Little variety of vegetation. Streambank dominated by one type of vegetation (trees, shrubs, flowering plants and grasses).				What did you see?			
Determine right/left bank by facing downstream									Did you see any nonnative vegetation? Check here if YES <input type="checkbox"/>			
Left bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	Score (Add both banks)
Right bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	
<b>10. Riparian Vegetative Zone Width</b>												
<b>What is the amount of buffer available?</b>	Buffer present; a large variety of vegetation extends at least three channel widths on each side.	Some buffer present; some variety of vegetation extends two to one channel width on each side. Human activities have impacted buffer zone.			Little or no buffer present; vegetation extends less than one channel width on each side. Human activities substantially impact buffer zone.				What did you see?			
Determine right/left bank by facing downstream									Did you see any nonnative vegetation? Check here if YES <input type="checkbox"/>			
Left bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	Score (Add both banks)
Right bank	5	4.5	4	3.5	3	2.5	2	1.5	1	.5	0	

Stream Habitat Score: Excellent (69-90) Good (46-68) Fair (23-45) Poor (0-22) Total second side \_\_\_\_\_

Please submit data at: [www.NCaquaticdatahub.org](http://www.NCaquaticdatahub.org)

Total first side \_\_\_\_\_

(Field form courtesy of Georgia Adopt-A-Stream Program)

Total \_\_\_\_\_



## BAU Habitat assessment



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### HABITAT ASSESSMENT GUIDELINES

March 2009 Revision

The North Carolina Division of Water Quality Biological Assessment Unit (BAU) developed an assessment technique most applicable to North Carolina streams. The habitat assessment protocol is based on best professional judgment of 8 habitat metrics including analysis of channel modification, four instream habitat measurements, one streambank measurement, and two riparian zone measurements. Scores are given for each of the eight metrics (seven for coastal plain and sandhills streams) and are then totaled (100 points possible).

Streams, or monitoring stations, within major ecoregion types and size categories can be compared to one another and to reference locations. However, at this time the habitat information functions primarily as a narrative evaluation. Habitat scores below 65 for the Mountain/Piedmont form are considered low to poor quality, while score 65 or higher are considered moderate to high quality.

This assessment is easy to perform, rapid, and can be relatively accurate with proper training. However, since it is a visual method of estimating overall habitat conditions, it is qualitative and can be subjective. Documentation of habitat characteristics at a sampling site can identify limiting factors that might affect biological communities.

Habitat assessment provides baseline information on stream conditions so that changes resulting from natural or human causes can be identified or predicted. Habitat assessments can also determine the consequences on the biota of alteration of stream conditions, such as land use changes and channelization. The goal is to relate habitat assessment to benthic macroinvertebrate or fish community data to give an overall condition of the biotic integrity of the stream. Annual training should be conducted in January-March of each year to maintain consistency between evaluators. This training will consist of independent scoring by all staff of sites with both low and high quality and discussion about differences in scores until consensus is reached on how a particular metric should be scored. Those clarifying discussions will be added to subsequent revisions of these guidelines.

## GENERAL PROCEDURES

- The habitat assessment is performed on the same reach from which the biological sampling is conducted, preferably a 200m of stream (100m minimum). Training sessions have revealed that the 100m minimum reach sometimes gave very different habitat scores between fish and benthos sampling when stream habitat varied greatly within 200m, but not within 100m. A 200m reach would minimize such differences, when habitat is variable. Complete the sampling first in order to get a close look at the habitat features.

The stream segment which is assessed should represent average stream conditions. The question was raised about whether we are sampling maximum habitat or average habitat, and so whether the sample reach represents average conditions. Benthic sampling does look for the best available habitat, but the change to a 200m habitat evaluation reach should give a representation of overall habitat.

- One person typically completes the Habitat Form, but should consult with other biologists about scoring atypical or unusual features at a site. The 2006 training noted the value of having a second person take a couple of minutes at the site to review the scores and reach consensus on any differences.
- When filling out the form, select the description which best fits the observed habitat and circle the score. If the observed habitat falls in between two descriptions, write in an intermediate score.
- There are eight different metrics in this index and a final habitat score is determined by adding the results of the different metrics. The form provides lines to subtotal individual metrics, have page subtotals and a final score which must be transferred to the front page.

## CAUTIONARY NOTES

Discussions in 2009 again expressed concern about seasonal changes in habitat scoring, and how high or low water could affect scoring for riffles and pools. The need for quarterly habitat evaluation at select sites to evaluate this concern was raised. The decision was again made to evaluate the habitat as is, not how it might look.

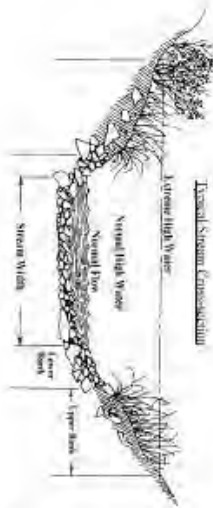
Some larger river sites might be evaluated with Piedmont biological criteria because their watershed is mainly Piedmont, but lie in Coastal Plain and so would need CP habitat form to be used. Site topography and substrate should outweigh map lines for determining which form to use.

You will find a more in-depth guide to scoring each parameter, including what to look for, why is it important, how to score the parameter and a definition of terms on [NCAquaticDatahub.org](http://NCAquaticDatahub.org).



**Supplement for Habitat Assessment Field Data Sheet**

Diagram to determine bank angle:



This slide is 45° bank angle.

Site Sketch:

Other comments:

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**Habitat Assessment Field Data Sheet**  
**Mountain/Piedmont Streams**

**TOTAL SCORE**

**Biological Assessment Branch, DWR**  
Directions for use: The observer is to survey a minimum of 100 meters with 200 meters preferred stream, preferably in an upstream direction, starting above the bridge pool and the road right-of-way. The segment which is assessed should represent average stream conditions. To perform a proper habitat evaluation the observer needs to get into the stream. To complete the form, select the description which best fits the observed habitats and then circle the score. If the observed habitat falls in between two descriptions, select an intermediate score. A final habitat score is determined by adding the results from the different metrics.

Stream \_\_\_\_\_ Location road: \_\_\_\_\_ (Road Name) \_\_\_\_\_ County \_\_\_\_\_

Date \_\_\_\_\_ CCR \_\_\_\_\_ Basin \_\_\_\_\_ Subbasin \_\_\_\_\_

Observ(s) \_\_\_\_\_ Type of Study:  Fish  Benthos  Basinwide  Special Study (Describe) \_\_\_\_\_

Latitude \_\_\_\_\_ Longitude \_\_\_\_\_ Ecoregion  MT  P  State Help  House Basin

Water Quality: Temperature \_\_\_\_\_ °C DO \_\_\_\_\_ mg/l Conductivity (corr.) \_\_\_\_\_ µS/cm pH \_\_\_\_\_

Physical Characterization: Visible land use refers to immediate area that you can see from sampling location - include what you estimate driving thru the watershed in watershed land use.

Visible Land Use: \_\_\_\_\_ %Forest \_\_\_\_\_ %Residential \_\_\_\_\_ %Active Pasture \_\_\_\_\_ %Active Crops  
 \_\_\_\_\_ %Rural Fields \_\_\_\_\_ %Commercial \_\_\_\_\_ %Industrial \_\_\_\_\_ %Other - Describe \_\_\_\_\_

Watershed land use:  Forest  Agriculture  Urban  Animal operations upstream

Width (meters) Stream \_\_\_\_\_ Channel (at top of bank) \_\_\_\_\_ Stream Depth (m) Avg \_\_\_\_\_ Max \_\_\_\_\_  
 Width variable  Large river >25m wide

Bank Height (from deepest part of riffle to top of bank-first flat surface you stand on): (m) \_\_\_\_\_

Bank Angle: \_\_\_\_\_ ° or  NA (Vertical is 90°, horizontal is 0°. Angles >90° indicate slope is towards in-channel, <90° indicate slope is away from channel, NA if bank is too low for bank angle to matter.)  
 Channelized Ditch  Both banks undercut at bend  Channel filled in with sediment  
 Deeply incised-steep, straight banks  Bar development  Bar development  Exposed bedrock  
 Recent overbank deposits  Heavy filamentous-algae growth  Green algae  Sewage smell  
 Excessive periphyton growth  Heavy filamentous-algae growth  Green algae  Sewage smell  
 Manmade Stabilization:  No  Y  R  Riprap, cement, gabions  Sediment/grate-control structure  Bank/levee  
 Flow conditions:  High  Normal  Low  
 Turbidity:  Clear  Slightly Turbid  Turbid  Milky  Oiled (from dyes)  
 Good potential for Wetlands Restoration Project?  Yes  No  Needs \_\_\_\_\_

**Channel Flow Status**

Useful especially under abnormal or low flow conditions.  
 A. Water reaches base of both lower banks, marginal channel substrate exposed.   
 B. Water fills >75% of available channel, or >5% of channel substrate is exposed.   
 C. Water fills 25-75% of available channel, many logs/slogs exposed.   
 D. Knows runs out of water.   
 E. Very little water in channel, mostly present as standing pools.

Weather Conditions: \_\_\_\_\_ Photos:  N  Y  Digital  S

Remarks:

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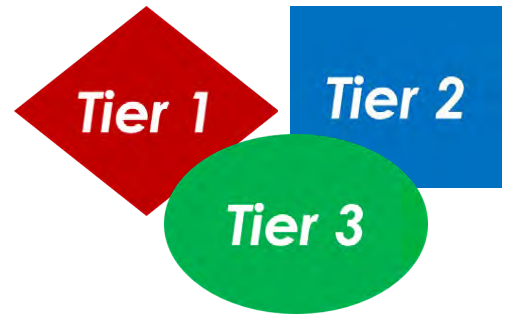
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# Chemical/Physical Protocols



## Collecting a water sample



From the New River Conservancy NRW methods manual

### Directly from the Source

When sampling from a small stream or river, it is appropriate to sample from the center of the stream/creek. Approach the stream from downstream, and travel upstream to your site. To avoid contamination, thoroughly rinse the water sample containers twice with the water to be sampled, discarding the rinse water downstream of your sampling location. If you are monitoring more than one site on the same stream, run tests on downstream site, then upstream site so as not to alter results.

### Using a bucket to collect sample water

If you are collecting from the streambank or bridge, try to wade in or throw bucket out as far as you safely can into the main channel. Try not to disturb the bottom of the water.

1. Using the water to be sampled, rinse the bucket twice downstream of the actual sampling location.
2. Then, gently lower the bucket into the water to avoid splashing and fill it about 2/3 full.
3. Once the sample is collected, be careful not to aerate or jostle the sample.



Quickly move on to the other monitoring tests to minimize the time between sample collection and measurement of parameters.



## Grab samples for lab analysis



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### Grab Samples

Grab samples are used to characterize the water at a particular point in time. All grab samples must be taken at approximately 6 inches (0.15 m) below the surface unless otherwise requested. Grab samples at a site should also be collected over a period of time not exceeding 15-minutes, in accordance with the DWQ Intensive Survey Unit SOP (DWQ, 2011)6. Collect grab samples in the same location where field parameters are taken. This should be in a region of well-mixed flow, as described in the section on “Sampling Location”, page 4). It is recommended that monitors wear a new pair of disposable laboratory gloves at each site to avoid sample contamination.

The following parameters are to be collected as grab samples in the coalition program, unless otherwise specified:

- Fecal coliform and E.coli
- Nutrients: ammonia (NH<sub>3</sub>); total Kjeldahl nitrogen (TKN); nitrate/nitrite (NO<sub>3</sub>+NO<sub>2</sub>); total phosphorus (TP)

NOTE: At sites where chlorophyll a is collected as a photic zone sample, nutrients will also be collected as a photic zone sample. At sites where chlorophyll a is sampled in summer months only, nutrients are to be collected as a photic zone sample year-round.

- Suspended residue (a.k.a. total suspended solids; TSS)
- Turbidity
- Metals: aluminum (Al), arsenic (As), cadmium (Cd), copper (Cu), chromium (Cr), iron (Fe), mercury (Hg), manganese (Mn), nickel (Ni), lead (Pb), zinc (Zn)



## GRAB SAMPLE PROCEDURES - DIRECT SAMPLING METHOD

- 1.) Always ensure bottle labels match the sampling site location.
- 2.) Remove the cap from the bottle just before sampling. Protect the bottle and cap from contamination. Avoid touching the inside of the bottle or cap. If the inside of the bottle or cap is touched, use another one. If the cap must be set down, place it so that the inside of the lid faces upward.
- 3.) Plunge the bottle into the water with the mouth facing downward and pointing upstream while avoiding surface scum. Take care to avoid dumping preservative out. The mouth should also be oriented away from the hand of the collector, the shore, the side of the sampling platform, or the boat.

Figure 1 Grab sample bottle orientation

- 4.) Once underwater, tip the bottle slightly upwards to allow air to exit and the bottle to fill.  
The mouth of the bottle should be approximately 6 inches (0.15 m) below the water surface.
- 5.) Once full, recap the bottle. Unless otherwise specified by the analytical laboratory, leave a small headspace. This allows the sample to be shaken prior to analysis.
- 6.) Add preservatives if needed.
- 7.) Ensure the bottle is labeled appropriately. Check that the following information is correct: site location, date, time, collector, parameter(s), and preservative(s).
- 8.) Place bottles in a cooler, follow the guidelines in regarding thermal preservation

## GRAB SAMPLING - INTERMEDIATE SAMPLING DEVICES

These devices are any type of sampling device that holds the sample prior to pouring it into a sample bottle, and are used when sampling from a bridge or area that the water cannot be reached. Intermediate sampling devices include cage samplers (Figure 2), weighted bottle frames, and similar custom devices.

Figure 2. Cage sampler



## GRAB SAMPLING - INTERMEDIATE DEVICES TECHNIQUE

1. Always ensure bottle labels match the sampling site.
2. Place the bottle(s) securely in the intermediate sampling device. Confirm that each bottle is held securely.
3. Remove the bottle cap(s) and lower the device to the water. Protect the inside of the bottle and cap from contamination. Avoid touching the inside of the bottle or cap. If the inside of the bottle or cap is touched, use another one. If the caps must be set down, place so that the inside of the lid faces upwards.
4. Swing the sampling device downstream and then allow it to drop into the water while pulling on the rope so as to position the bottle openings upstream. Take care not to disturb the bottom sediment.
5. Allow bottle(s) to fill.
6. Pull the sampling device out of the water.
7. Take care not to dislodge dirt or other material from the sampling platform.
8. Unless otherwise specified by the analytical laboratory, leave a small headspace; this allows the sample to be shaken prior to analysis.
9. Add preservatives if needed
10. Recap the bottle(s), remembering not to touch the inside of the bottle or cap.
11. Ensure each bottle is labeled appropriately. Check that the following information is correct: site location, date, time, collector, parameter(s) and preservatives(s).
12. Place bottles in a cooler, on ice, within 15 minutes for transport to the lab. Follow the guidelines in the section “Bottles and Preservation” (page 31) regarding thermal preservation of samples on ice.

### 4. Photic Zone Sampling

Photic zone samples, also known as depth-integrated composite samples, are collected in the portion of the water column where photosynthesis by phytoplankton occurs. The photic zone is defined by the DWR as twice the Secchi depth. Photic zone samples are collected by lowering an integrated depth-sampling device, such as a Labline® sampler to twice the Secchi depth and then slowly raising the device to the surface to obtain a representative water sample. The sampler is raised and lowered at a slow, constant pace throughout the region of twice Secchi depth until the sampler is full.

Photic zone samples are collected for:

- Chlorophyll a samples (at designated sites, assuming adequate water depth)
- Nutrients (to be collected as photic zone samples at sites where chlorophyll a is collected as photic zone samples. Nutrients should be collected year round as photic zone samples even when chlorophyll a is only collected in the summer months)

Figure 4. Labline® sampler for photic zone measurements

Photic Zone Sampling Technique



To collect a vertical spatial composite sample in the photic zone:

- 1.) Measure and record the Secchi depth (See “Secchi Depth Measurements”, page 14).
  - 2.) Prior to collecting sample for analysis, rinse the integrated sampling device (Labline®) with sample water:
    - a. Lower the Labline to the water. Take care not to disturb sediment.
    - b. Fill the Labline with water with water from the photic zone.
    - c. Raise to surface, swirl, and pour out water. Pour water away from sampling location so that the rinse water will not disturb the sample. After rinsing, collect the sample for analysis.
  - 3.) Lower the Labline to twice the Secchi depth. (i.e. if the Secchi depth is 0.5 m, lower the Labline so that the fill hole on the sampler is at 1 m depth).
  - 4.) Raise the sampler to the surface at a slow, constant rate so as to collect a representative sample of the water column in the photic zone.
  - 5.) Continue to lower and raise the Labline throughout the photic zone in a slow, constant motion until the sampler is full. Maintain a steady pace so as to ensure a representative sample.
  - 6.) Pour sample from Labline into appropriate sample bottle.
- NOTE: Chlorophyll a sample bottles must protect the sample from light. Brown opaque bottles are commonly used.
- 7.) Rinse Labline thoroughly with distilled water after using to clean between sites.



## pH: Colorimetric Kit



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From the New River Conservancy NR Water Watchers methods manual

**Equipment:** Lamotte pH kit

**Method:**

1. Rinse the test tube and cap twice and toss rinse water downstream of your sampling location.
2. Fill the sample test tube to the 10 mL with the sampling water. The bottom of the meniscus should be even with the line. Use plastic dropper to adjust water level in test tube.
3. Add ten drops of the wide range indicator while holding the reagent bottle completely upside down
4. Cap the test tube and gently mix the sample ("rainbow shake").
5. Insert the test tube in the Octa-slide Viewer next to the slot that most closely matches the sample tube color. Record the number as the pH value.



When the color observed is between 2 colors on the comparator, the value is reported to the nearest 0.5 unit.



## pH: Test Strips



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### From the CMC non-tidal methods manual

#### GATHERING MATERIALS AND EQUIPMENT LIST

- Fisher pH strips (0-14)
- Sampling pole (if using)
- Bucket (if using)

#### CHECKING YOUR EQUIPMENT BEFORE GOING OUT IN THE FIELD

- Check your strips to make sure that they are not discolored or have been previously dampened.

#### I. Collecting directly in stream

1. Always proceed upstream to allow the flow of the water to push any disturbed sediment downstream of where you will be collecting the sample.
2. Be sure any sediment or debris disturbed from your movement in the streambed is not present where you will collect the sample.
3. Carefully remove strip from the box and close the box when not in use.
4. Dip the strip in the water until fully moistened.
5. Allow for colors to fully develop about 1 to 2 minutes.
6. Compare your strip to the chart provided.
7. Record your value to the nearest 0.5 units.
8. Properly dispose of your strip when you are finished.

## II. Collecting with a bucket

1. Throw the bucket out as far as possible in the main channel, and try not to disturb the bottom.
2. Rinse the bucket three times with stream water collected downstream of your sampling location.
3. Fill the bucket with the sample water to 3/4 full.
4. Carefully remove strip from the box and close the box when not in use.
5. Dip the strip in the water until fully moistened.
6. Allow for colors to fully develop about 1 to 2 minutes.
7. Compare your strip to the chart provided.
8. Record your value to the nearest 0.5 units.
9. Properly dispose of your strip when you are finished.
10. Mark on your data sheet that the measurement was taken from a bucket.

## EQUIPMENT STORAGE

1. Store your strips in a cool dark place.



## pH: Pen Meter

**Tier 2**

**Tier 1**

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### From the CMC non-tidal methods manual

Prior to sampling, calibrate the probe according to the manufacturer's instructions

### Method (with bucket):

1. Throw the bucket out as far as possible in the main channel, and try not to disturb the bottom.
2. Rinse the bucket three times with stream water collected downstream of your sampling location.
3. Fill the bucket with the sample water to 3/4 full.
4. Place your probe in the bucket of water and swirl gently. Allow the reading to stabilize.
5. Record your reading on your data sheet. Mark on your data sheet that the measurement was taken from a bucket.

### Method (no bucket) :

1. Always proceed upstream to allow the flow of the water to push any disturbed sediment downstream of where you will be collecting the sample.
2. Be sure any sediment or debris disturbed from your movement in the streambed is not present where you will collect the sample.
3. Place your probe 0.3 m (about one foot) beneath the surface of the water.
4. Wait for the probe to stabilize and record your reading on your data sheet.
5. Turn off your probe and replace the protective cap.
6. Record your reading on your data sheet and the measurement depth.

## Both methods: post-sampling calibration

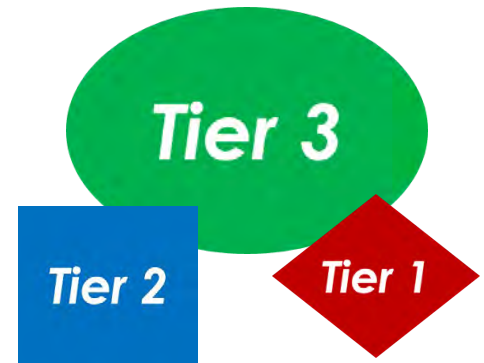
To ensure the probe has maintained proper calibration, it is important to verify no significant probe drift has occurred. The procedures listed below will verify the probe did not drift outside QA/QC specifications. DO NOT CALIBRATE the probe during this check. Doing so will invalidate the data collected during the sample run.

1. Rinse off the probe and probe tip with distilled water and wipe dry using a soft cloth. Washing the probe will remove any material that may reduce probe life.
2. Place the probe into a container of pH 7.00 buffer. You may use the same buffer used during the morning calibration as long as the buffer was covered and appears clean.
3. Allow the probe to stabilize and record the temperature and pH reading in the end of day temperature in °C.
4. Rinse the probe and repeat the end of day check process using the 4.00 or 10.00 buffer.





## pH: Probe



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### From the DWR Intensive Survey Branch (ISB) SOP

4.1.1. Precision and accuracy:  $\pm 0.2$  pH unit represents the limit of accuracy under normal conditions for measurements of water and poorly buffered solutions. For this reason, report pH values to the nearest 0.1 pH unit. Calibrate instrument within 0.2 pH units of the standard pH buffer value.

4.1.2. Calibration Reagents - Calibrate the electrode system against standard buffer solutions of known pH. Always use fresh commercially made buffers to calibrate field meters. Buffer solution and samples should be stored in polyethylene bottles. Never pour decanted or used buffer solution back into the original bottle.

4.1.3. Procedure - Always follow the manufacturer's instructions for pH meter storage and preparation of electrodes. Recommended short-term storage of electrodes varies with type of electrode and manufacturer. Never store probes in deionized water; tap water or pH buffer 4.0 is preferred. Note: All field meters should be calibrated before and checked after sampling activities daily. The calibration data should be entered on a meter calibration sheet.

### 4.2. Multiparameter YSI or Hydrolab Meters

The Hydrolab and YSI meters used by ISB all have the same basic method for calibration.



## Water/air temperature: Handheld Thermometer

Tier 2

Tier 1

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From the New River Conservancy NRW methods manual

**Equipment:** armored or digital thermometer

Temperature is reported in degrees Celsius ( $^{\circ}\text{C}$ ). The table in Appendix 1 converts Fahrenheit to Celsius. Always measure air temperature before water temperature.

### Air Temperature Measurement

#### Method:

1. Locate a place near your site and hang the thermometer out of the direct sun.
2. Wait 3-5 minutes to allow the thermometer to equilibrate. (You can begin filling out page 1 of the datasheet while you wait for the thermometer to equilibrate.)
3. Record air temperature to the nearest **0.5  $^{\circ}\text{C}$**  on the datasheet.

### Water Temperature Measurement

#### Method (with bucket):

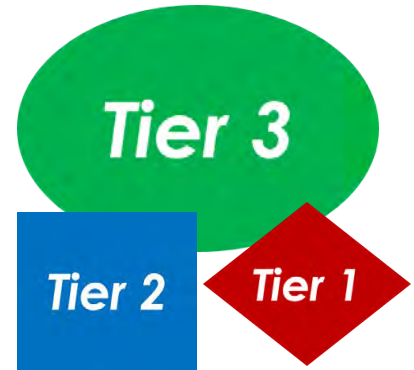
1. Hang the thermometer in the bucket while holding the thermometer from the top with the bulb or metal part submerged in the water, not letting the digital head get wet.
2. Wait for the numbers to stop changing, letting the thermometer equilibrate. (but not long enough for water temperature to change)
3. Record water temperature to the nearest 0.5  $^{\circ}\text{C}$ .

#### Method (no bucket):

Hold the thermometer with the bulb or metal part submerged directly in the stream, following steps 2 and 3 above.



## Water/air temperature: Probe



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### From the DWR Intensive Survey Branch (ISB) SOP

Temperature measurements are taken by a multiparameter meter (Hydrolab or YSI) or dial Celsius-thermometer or a thermistor. Below are some general considerations while collecting water temperature data.

- The meter should have a scale marked for every 0.1°C.
- Allow the multiparameter meter or thermometer to rest in the water long enough to permit equilibrium.
- Temperature sensors on the Hydrolab and YSI meters are factory set and do not require recalibration.
- At least once a year check the meter thermometer against a precision thermometer certified by the National Institute of Standards and Technology (NIST).
- Temperature readings must be record as degrees Centigrade (°C) to the nearest tenth of a degree. During field use, the temperature readings should always be read when they are stable and before the other parameters are read to ensure stable readings for all parameters.



## Fecal coliform: Coliscan Easygel

Tier 2

Tier 1

---

### From the New River Conservancy NRW methods manual

#### Before going to the field:

Perform the following equipment preparations the evening before sampling or as indicated below:

1. Take bacteria media solution bottle (1 sample per site) out of freezer and put in refrigerator to thaw. If you do forget to thaw your media, fill a bowl with hot water and let the frozen media solution sit until liquefied.
2. Place ice pack in freezer so it will be cold for the next day.
3. Before going out into the field, put ice pack and sterile sample bottle in the cooler.
4. If using an incubator prepare incubator: Turn on a few hours ahead of sampling time to maintain incubator at 37°C (= 98.6°F).

#### In the field: Bacteria Sample Collection

1. Collect the water sample.

If collecting by wading: Wade into the main flow of the stream; take a few steps upstream with minimal disturbance; then reach upstream away from your body to collect the sample.

If collecting by bucket: Make sure not to touch inside of bucket with your hands. Throw the bucket out as far as possible in the main channel, and try not to disturb the stream bottom. Rinse the bucket twice with stream water collected downstream of your sampling location. Fill the bucket with the sample water to 2/3 full.

Cap the sample bottle and immediately place the sample bottle on ice in cooler.

**NOTE:** Be careful not to let the bottle lid touch anything to prevent sample contamination.

If collecting samples from more than one site, label each sample bottle with the site designation using a permanent waterproof marker.

## After returning from the field:

### Bacteria Sample Plating

If collecting samples from more than one site write the site designation, sample #, date, and time on the bottom (the smaller, taller piece) of the Petri dish with a permanent marker.

1. Record the expiration date of the media bottle on your datasheet.
2. Use proper technique to keep pipette sterile: open pipette packet bulb-side first so that you do not contaminate the tip.
3. Pipette the desired volume (1.0 – 5.0 milliliters) of sample water directly into Coliscan media bottle, carefully not to touch the pipette to the bottle.

**NOTE:** Be careful not to let the bottle lid touch anything to prevent sample contamination.

4. Gently swirl (do not shake) bottle of Coliscan media containing the sample water.
5. Pour the entire contents into a Petri dish.

**NOTE:** Only open the Petri dish long enough to pour in the sample.

6. Gently swirl Petri dish so the Coliscan media covers the entire bottom. For safety purposes, tape the Petri dish shut at this point.

7. Allow the media to solidify for approximately 60 minutes prior to incubation. (Amount of time will vary based on room temperature.)

8. Incubate plated Petri dish(es) upside down. If using an incubator try to maintain at 37°C (= 98.6°F) for 24 hours or if not, place the Petri dish(es) in the safest, warmest spot you can find. Depending on temperature, the plates may take 48-72 hours for colonies to form.

9. Record the average incubator or room temperature on the datasheet as well as the # of hours that the plate(s) were "incubated".

**NOTE:** As soon as plates are removed from incubator, they must be scored.

### Bacteria Scoring

1. Place the Petri dish (es) on a white background or in natural sunlight. Count the number of **dark blue** (NOT TEAL) to **purple** (NOT PINK) colored colonies larger than pinprick size on each plate. Do not pay attention to halos around the dots, but **only the center color**.
2. Record this number in the column labeled "Total # of purple or dark blue colonies on plate" on the data form.

### Bacteria Cleanup and Disposal

1. Throw used pipette in the trash.
2. Rinse empty Coliscan bottle once with tap water and dispose of in the trash. (If media bottle is not rinsed, pathogens could grow in the remaining media.)
3. Add bleach or rubbing alcohol to the Petri dish to completely cover the solid media. Allow dish to stand for at least 10 minutes to ensure all bacteria have been killed.
4. Place the plate in a zip-lock bag and dispose of in the trash.

**NOTE:** If you conduct the sampling procedures away from home or on a boat, you need a special container for safe disposal of the test samples. A plastic milk jug or jar works well and is easy to obtain. Fill this container about  $\frac{1}{2}$  to  $\frac{3}{4}$  full with kitty litter to absorb the moisture. When the litter is saturated, place the closed jar in double plastic garbage bags and dispose of in the trash.



## Fecal coliform: Grab samples for lab analysis

Tier 3

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### From the DWR Intensive Survey Branch SOP

- a. Collect sample with a 250 ml wide-mouth sterile plastic bottle supplied by the DWR Laboratory. These bottles must contain sodium thiosulfate and EDTA reagents.
- b. Coliform sample is always collected as a surface grab sample. In no case should composite samples be collected for microbiological examination.
- c. Do not rinse bottle with sample, but fill it directly to within 1-2 inches from the top to allow mixing of the sample before analysis.
- d. Use caution to avoid contaminating the sample with fingers, gloves, or other materials.
- e. Cool to 4°C and return to lab in less than 6 hours from time of collection. DWR's or a state certified lab will analyze any coliform samples that are received in less than 24 hours; however, the data may not be acceptable for some uses due to extended holding time.

### IV. WATER SAMPLE COLLECTION AND PRESERVATION

#### 2.3.2. Surface Sampling By-Hand

- a. Grab sample should be collected directly into the sample bottle.
- b. Remove the bottle top to protect bottle and cap from contamination; avoid touching the inside of the bottle and cap.
- c. Grasp the bottle securely near the base with one hand and plunge the bottle mouth down into the water to avoid surface scum. Position the bottle towards the current flow and away from the hand of the collector, the shore, the side of the sampling platform, or boat. The sampling depth should be 0.15 m (about 6 inches) below the water surface.
- d. If the water body is static, create an artificial current by moving the bottle away from the sampler while tipping the bottle slightly to allow water to enter.
- e. Tip the bottle slightly upwards to allow air to exit and the bottle. Fill the bottle to within 1-2 inches of the top.
- f. After removal of the bottle from the stream, tightly stopper and label the bottle.

### 2.3.3. Surface Sampling by Weighted/Cage Bottle Frame

- a. Remove the cover and lower the device to the water.
- b. It is preferable to use nylon rope which does not absorb water and will not rot.
- c. Swing the sampling device downstream and then allow it to drop into the water while pulling on the rope so as to direct the bottle upstream.
- d. Pull the sample device rapidly upstream and out of the water, simulating the scooping motion of grab sampling.
- e. Take care not to dislodge dirt or other material from the sampling platform.





## Dissolved Oxygen: CHEMetrics ampoules



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From the Hoosier Riverwatch Volunteer Stream Monitoring Training Manual

**Equipment:** CHEMetrics K-7512 Dissolved Oxygen Test Kit

1. Fill the sample cup to the 25 mL mark with the sample to be tested
2. Place the ampoule, tip first, into the sample cup. Snap the tip. The ampoule will fill, leaving a bubble for mixing
3. To mix the ampoule, invert it several times, allowing the bubble to travel from end to end.
4. Dry the ampoule and wait 2 minutes for color development.
5. Obtain a test result by placing the ampoule between the color standards until the best color match is found
6. Record the dissolved oxygen concentration to the nearest mg/L. Rinse the glass tip out of sample cup into a waste container.

Note: The CHEMetrics ampoules and color standards contain a reagent which deteriorates upon prolonged exposure to light. They will remain stable only if stored in the dark. The reagent should be a light straw color with no hint of blue or green when the ampoule is removed from the box. The normal shelf life of the color standards is two years.



## Dissolved Oxygen: Pen meter

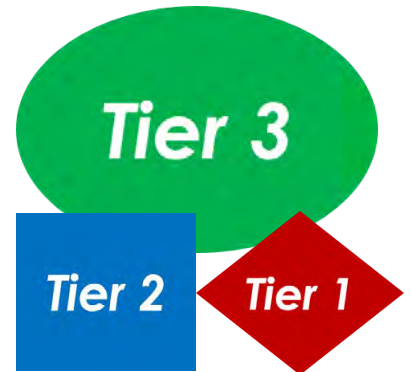
**Tier 2**

**Tier 1**

- 
1. Inspect dissolved oxygen (DO) meter for damage - repair as necessary.
  2. Rinse probe and cable with Distilled or Deionized water.
  3. Prepare probe and DO meter in accordance with instrument manufacturer's operating procedures. Make certain probe contains sufficient electrolyte and the oxygen sensor membrane is in good repair.
  4. Calibrate probe and meter using the fresh water - air calibration method. Correct calibration value for temperature and altitude; adjust meter accordingly.
  5. When possible place probe directly into the stream, or water to be measured. If not possible, place probe into beaker filled with sample. Manually raise and lower probe through sample. Allow sufficient time for probe to stabilize to sample temperature and dissolved oxygen concentration.
  6. Read dissolved oxygen value. Record appropriate data on field forms.



## Dissolved Oxygen:probe



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From the CMC non-tidal water quality methods manual

### Gathering material and equipment list

- Various models of dissolved oxygen probes and meters
- Distilled or DI water
- Bucket (if using)

### Checking your equipment before going out in the field

- Examine the probe for wear or damage.
- Make sure there is sufficient battery life for your field trip and check for battery leaks.
- Make sure all openings are sealed tight.
- Calibrate your meter before each sampling day.
- Various models of dissolved oxygen probes and meters
- Distilled or DI water
- Bucket (if using)

### Calibration

With practice and proper care for the dissolved oxygen probe, users can complete the entire DO probe calibration process within 5-10 minutes.

NOTE: Some probes may differ in displaying values. For DO probes, parts per million (ppm), and milligrams per liter (mg/L) are the same value. In addition, barometric pressure may be displayed in millibars (mBar) or in millimeters of mercury (mmHg).

Calibrations before field sampling must be performed to standardize the response of each probe. Record calibration in logbooks for each instrument and/or sensor. The logbooks document all calibration,

maintenance, and servicing information. Calibrations before field sampling should be performed indoors. Allow the probes to stabilize to room temperature. Follow all manufacturer specifications for calibration and maintenance

### At the stream

NOTE: Replicate tests are taken to guard against error. Don't forget to measure twice!

#### Collecting with a bucket - from a dock, bridge, or boat

1. Throw the bucket out as far as possible in the main channel, and try not to disturb the bottom.
2. Rinse the bucket three times with sample water collected downstream of your sampling location.
3. Fill the bucket with the sample water to 3/4 full.
4. Place your probe in the bucket of water and swirl gently. Allow the reading to stabilize.
5. Record your reading on your data sheet. Mark on your data sheet that the measurement was taken from a bucket.

#### Measuring directly in the stream

1. Place your probe 0.3 m beneath the surface of the water.
2. Wait for the probe to stabilize, and then record your reading and the depth at which you recorded your reading.

#### Wading - collecting directly in the waterway

1. Always proceed upstream to allow the flow of the water to push any disturbed sediment downstream of where you will be collecting the sample.
2. Be sure any sediment or debris disturbed from your movement in the streambed is not present where you will collect the sample.
3. Place your probe 0.3 m beneath the surface of the water.
4. Wait for the probe to stabilize and record your reading on your data sheet.
5. Turn off your probe and replace the protective cap.
6. Record your reading on your data sheet and the measurement depth.

### After sampling calibration check

After the sample run is complete, return the probe to the calibration station to perform a quick post check. The post check consists of placing the probe in the DO calibration chamber and letting it equalize. This may take between 2 to 10 minutes depending on the condition of the probe. Calibrations after field sampling must be performed to check the response of each probe. Record calibration in logbooks for each instrument and/or sensor. The logbooks document all calibration, maintenance, and servicing

information. Calibrations after field sampling should be performed indoors, but if performed outdoors, note that the dissolved oxygen value can be different than the pre-field calibration value.

Follow all manufacturer specifications for calibration and maintenance.

### **Equipment cleaning and storage**

1. Follow manufacturer's instructions for cleaning and storing the probe.
2. Ensure the probe is cleaned and well maintained. After each sample run, rinse off the probe with distilled water. Use a soft cloth and gently dry the probe and sensor.
3. Store the probe tip in the cap provided by the manufacturer.
4. If the calibration or end of day check indicates there is a problem with the probe, and standard cleaning does not produce acceptable results, replacement of the sensor cap may be necessary. Contact a Project Team Member to get a replacement sensor cap.
5. Store the probe in a clean, cool, and dry space.



## Water clarity: turbidity tube



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### From the New River Conservancy NRW methods manual

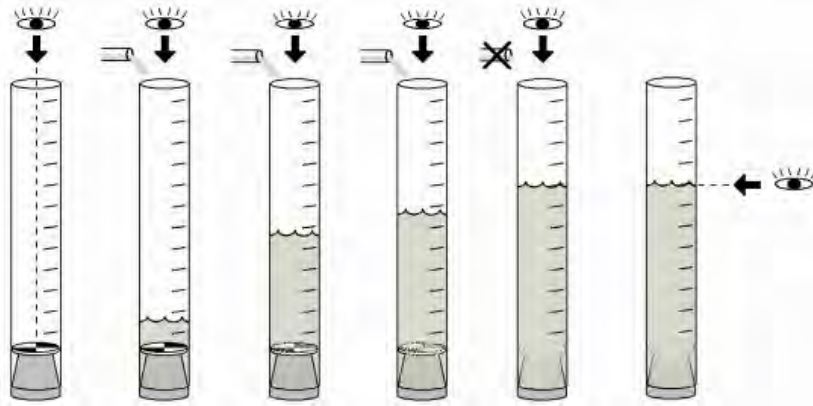
Turbidity tubes are a type of equipment used for measuring transparency of water in streams and rivers. They are helpful for measuring transparency in situations where the stream is too shallow for the Secchi disk to be practical and for running waters where flow is too fast that the Secchi disk cannot remain vertical. Sample water collected either directly from the stream or from the sampling bucket is analyzed.

**Equipment:** Turbidity tube. See Appendix 4 for instructions for construction of a tube and conversion of centimeters to NTU table.

**Method:**

1. Fill the turbidity tube with your sample water. Water may be collected directly from the stream in the vicinity of the sampling location if the stream is too small to fill the bucket, or sample water collected in the sampling bucket may be used. To collect water directly from the stream, point the top of the tube in the upstream direction and collect surface water, being careful not to disturb the streambed. To analyze water collected in the bucket, pour sample water from the bucket water directly into the transparency tube. Alternatively, collect water in a pitcher and pour into the tube.
2. Remove sunglasses if you are wearing them and stand with the sun to your back.
3. Look down through the opening of the tube. If the black and white pattern is not visible, pour a small amount of the water out. Look down through the opening of the tube again. Continue process until the black and white pattern at the base of tube faintly begins to appear.

4. Record the level of water remaining via the centimeter rule on the side of tube.





## Conductivity: Pen meter

Tier 2

Tier 1

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### Calibration:

Calibration is critical for accurate data collection. Use a calibration standard that is close to your intended sample value. The general steps are listed below. Follow the manufacturer's instructions.

1. Remove the cap and power on the unit.
2. Dip the sensor in at least 30mm of calibration standard.
3. Stir gently and press the calibration button to begin.
4. If the reading is within the calibration range of the automatically recognized standards the unit will report the value.
5. If the value matches the standard, accept the standard and finish the calibration.

**Taking a measurement:** The general steps are listed below. Follow the manufacturer's instructions.

1. Remove the cap and power on the unit.
2. If probe is a multi-meter, change mode to conductivity.
3. Dip sensor in at least 30mm of water quality sample.
4. Stir gently while reading stabilizes and wait for the icon to stop blinking.
5. Record the reading

*Many units shut off in 8-10 minutes of non-use to conserve batteries.*





## Salinity: Pen meter

Tier 2

Tier 1

---

### Calibration:

Calibration is critical for accurate data collection. Use a calibration standard that is close to your intended sample value. The general steps are listed below. Follow the manufacturer's instructions.

1. Remove the cap and power on the unit.
2. Dip the sensor in at least 30mm of calibration standard.
3. Stir gently and press the calibration button to begin.
4. Adjust the value to match the standard.
5. Accept the standard and finish the calibration.

**Taking a measurement:** The general steps are listed below. Follow the manufacturer's instructions.

1. Remove the cap and power on the unit.
2. If probe is a multi-meter, change mode to salinity.
3. Dip sensor in at least 30mm of water quality sample.
4. Stir gently while reading stabilizes and wait for the icon to stop blinking.
5. Record the reading

*Many units shut off in 8-10 minutes of non-use to conserve*



## Chlorophyll A: Grab samples for lab analysis

Tier 3

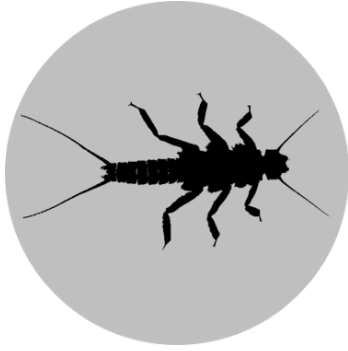
Tier 2

**From the NCDEQ-DWR Intensive Survey Branch SOP**

See [page 28 for grab sample collection guidance](#).

Chlorophyll a - Chlorophyll a is the photosynthetic green, photosynthetic pigment contained in plants. The measurement of this pigment provides an estimate of algal biomass.

Collection method: Use a 500 ml wide-mouth opaque plastic bottle to collect the sample. Cool to 4°C. Sample must be received by the laboratory in less than 24 hours.



## SOS Benthic macroinvertebrate assessment



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**From the Issac Walton League of America (IWLA) Save Our Streams (SOS)**

### **Biological Monitoring Instructions for Stream Monitors**

Surveying stream macroinvertebrates provides information about the health of your stream. Many stream-dwelling organisms are sensitive to changes in water quality. Their presence or absence can serve as an indicator of environmental conditions.

Monitoring should be conducted at the same station (location) each time you sample. If you want to monitor several stations on your stream, make sure the stations are no closer than one-quarter mile. This means, for example, that if you want to monitor a one-mile segment of a stream, you can have a maximum of four monitoring locations. If the stations are spaced more closely, the monitoring activity may become the main impact on water quality.

Carefully record the location of your monitoring station on your Biological Monitoring Data Form. Include roads, bridges, and significant landmarks. Use your smartphone's GPS functionality to determine your longitude and latitude.

### **THINGS TO CONSIDER**

If you are monitoring more than one station, begin monitoring downstream and move upstream. This will prevent macroinvertebrates disturbed by the first test from washing downstream and being captured in your net a second time. Each survey should record only the organisms present at that particular location and time.

When scheduling monitoring events, remember that excessive monitoring can become the major threat to stream health because each monitoring event disturbs the streambed and dislodges macroinvertebrates. In general, monitoring stations should have two months to recover from a monitoring event. It is crucial to the integrity of your data that you do not over-monitor your stations. There is some flexibility in this rule. For example, if an oil spill occurs, you might want to monitor your stream, even if you have done your six surveys for the year. The data you collect might be the only data available on the immediate impacts of the spill.

The methods described in these instructions are for use in wadeable streams. To be wadeable, the water level in the stream must not exceed the height of your knees. When planning monitoring sessions for younger people, please keep in mind that knee height varies greatly between adults and children.

There are two sampling methods available to collect aquatic macroinvertebrates. Muddy Bottom Sampling is used in streams that do not have riffles, i.e. a streambed feature with cobble-sized stones between 2 to 10 inches in diameter where the water bubbles over the rocks. If your stream has riffles, please refer to the Rocky Bottom Sampling section.

### **MUDDY BOTTOM SAMPLING**

The Muddy Bottom Sampling method is intended for volunteers sampling streams that do not have rocky bottoms or riffles. Muddy bottom streams are composed of muddy or sandy substrate, overhanging bank vegetation, and submerged woody and organic debris. This method enables sampling of streams where kick-seining techniques do not yield the best representative sample of macroinvertebrates or allow easy collection from the most productive aquatic habitats.

Monitoring is conducted using an aquatic D-frame or dip net with 1/32-inch mesh and a four-foot pole. The dip net is used to sample a wide variety of habitats and collect many different kinds of organisms.

Before you begin monitoring, familiarize yourself with the four main habitats that can exist along muddy bottom streams: steep banks/vegetated margins, silty bottom with organic matter, woody debris with organic matter, and sand/rock/gravel substrate. Search for these habitats along a 50-foot section upstream from the monitoring station.

### **MUDDY BOTTOM SAMPLING EQUIPMENT**

- One D-frame aquatic dip net with mesh of 1/32 inch
- “Field Guide to Aquatic Macroinvertebrates”
- Monitor’s Guide to Aquatic Macroinvertebrates
- Biological Monitoring Data Form
- Two small magnifier boxes (optional)
- Magnifying glass (optional)
- Shallow plastic pan
- Specimen jars or ice cube trays for sorting organisms
- One screen-bottom bucket with a mesh of 1/32 inch (optional)
- Tweezers or forceps
- Clipboard (optional)
- White sheet or plastic trash bag (optional)
- Old sneakers or sandals that secure to your feet. Waders may be preferred in cold weather or for additional leg protection when water is cloudy.

Following are simple descriptions of the habitat types and collection techniques for each habitat.

### **Steep banks/vegetated margins**

The area along the bank and the edge of the water body consists of overhanging bank vegetation, plants living along the shoreline, and submerged root mats. Vegetated margins may be home to a diverse assemblage of dragonflies, damselflies, and other organisms. Move the dip net in a bottom-to-surface motion, jabbing at the bank to loosen organisms. Each scoop of the net should cover one foot of submerged area.

### **Silty bottom with organic matter**

Silty substrates with organic matter can be found where the water is slow-moving and where there is overhanging vegetation or other sources of organic matter. The substrates harbor burrowing organisms such as dragonflies or burrowing mayflies. Collect samples by pushing the net upstream with a jabbing motion to dislodge the first few inches of organic layer.

### **Woody debris with organic matter**

Woody debris consists of dead or living trees, roots, limbs, sticks, and other submerged organic matter. It is a very important habitat in slow-moving rivers and streams. The wood traps organic particles that serve as food for the organisms and provides shelter from fish and other predators.

To collect woody debris, approach the area from downstream and hold the net under the section of wood you wish to sample, such as a submerged log. Rub the bottom of the net frame along the surface of the log for a total surface area of one foot. It also is good to dislodge some of the bark, as organisms may be hiding underneath. You can also collect sticks and leaf litter and rub roots attached to submerged logs. Be sure to thoroughly examine any small sticks you collect with your net before discarding them. There may be caddisflies, stoneflies, riffle beetles, and midges attached the bark.

### **Sand/rock/gravel substrate**

In slow-moving streams, bottoms are generally composed of only sand or mud because the water is not fast enough to transport large rocks. Sometimes you may find a gravel bar located at a bend in the river. The bottom can be sampled by pushing the net upstream with a jabbing motion to dislodge the first few inches of gravel, sand, or rocks. You may want to gently wash the gravel in your screen-bottom bucket and then discard the gravel into the stream.

To provide for accuracy of collection and comparability of data from one station to another, take a total of 20 scoops from the different habitats. Ideally, you should identify locations for all four habitat types and collect the following number of scoops from each:

- 10 scoops from steep banks/vegetated margins
- 3 scoops from silty bottom with organic matter
- 4 scoops from woody debris with organic matter
- 3 scoops from sand/rock/gravel substrate

If one of the habitat types is not present, divide the number of assigned scoops from that habitat between the other habitat types that are present. For example, if the stream does not have sand/rock/gravel substrate, take one extra scoop from each of the other three habitat types. The most important thing is to have a total of 20 scoops and to make sure all habitat types that are present are represented. The D-frame net is one foot wide, so one scoop equals one square foot being monitored. If you have large rocks (greater than two inches in diameter), it is important to dislodge any burrowing organisms. To do this, hold the net on the downstream side of the rocks. In a one-square-foot area in front of the net, gently kick up the rocks with your toes or push them free with your fingers. This should dislodge burrowing organisms and allow them to wash into your net.

After collecting some samples, dump the net into a shallow white pan filled with a few inches of water. Each time you sample, sweep the mesh bottom of the D-frame net back and forth through the water (not allowing water to run over the top of the net) to rinse fine silt from the net. This will avoid a large amount of sediment and silt from collecting in the pan and clouding the water.

Collect organisms from the net or pan and place them in similar groups as you go through the sample. This will make your identification quicker when you are ready to record results on your survey form. Plastic ice cube trays are helpful when sorting the sample. For example, put all organisms with two tails in one section and all organisms with three tails in another section. See the “Identification” section for details on identifying the organisms in your sample.

### **ROCKY BOTTOM SAMPLING**

The Rocky Bottom Sampling method is intended for volunteers sampling streams that have rocky bottoms or riffles. A kick-seine net – a finely meshed net with supporting poles on each side – is the best tool to use for collecting macroinvertebrates in rocky bottom streams. The Rocky Bottom Sampling method uses a kick-seine net that is 3-feet square with 1/16- or 1/32-inch mesh. Both sizes capture the full range of macroinvertebrate species included in this monitoring method. However, the 1/32-inch mesh net will provide you with a larger sample because it captures younger, and therefore smaller, organisms of each species, and some state and local government agencies require use of the 1/32-inch mesh.

## ROCKY BOTTOM SAMPLING EQUIPMENT

- Kick-seine
- “Field Guide to Aquatic Macroinvertebrates”
- A Guide to Aquatic Insects and Crustaceans
- Biological Monitoring Data Form
- Two small magnifier boxes (optional)
- Magnifying glass (optional)
- Shallow plastic pan
- Specimen jars or ice cube trays for sorting organisms
- Tweezers or forceps
- White sheet or plastic trash bag (optional)
- Clipboard (optional)
- Camera (optional)
- Squirt bottle (optional)
- Glass vials for collecting macroinvertebrate samples and 70-percent alcohol for specimen preservation (optional)
- Old sneakers or sandals that secure to your feet. Waders may be preferred in cold weather or for additional leg protection when water is cloudy.

Select a riffle that is a shallow, fast-moving area of water with a depth of 3 to 12 inches and cobble-sized stones (2 to 10 inches) or larger. Before entering the stream, record observations about riffle composition on the back of the Biological Monitoring Data Form.

For the smaller rock sizes, remember that silt feels like talcum powder and sand feels gritty. If your riffle had 40 percent silt, 10 percent gravel, and no cobbles, you should either find another station to monitor or switch to the Muddy Bottom Sampling method.

Place the kick-seine net at the downstream edge of the riffle. Use rocks to secure the net tightly against the streambed so that no organisms escape under the net.

Don't allow any water to flow over the top of the net either – organisms can escape over the net. Also, if water is flowing over the top of the net, the water level is too high for safe monitoring. Monitor the streambed for a distance of three feet upstream of the kick-seine and across the width of the net.

Firmly and thoroughly rub your hands over all rock surfaces to dislodge any attached insects. After you have rubbed off any macroinvertebrates, carefully place each large rock outside of your three-foot sampling area. Stir up the bed with your hands and feet until the entire area has been searched. All exposed and detached organisms will be carried into the net. Then, for at least 60 seconds, use the toe of your shoe to jab the streambed with a shuffling motion, moving towards the net.

Disturb the first few inches of sediment to dislodge burrowing organisms.

Before removing the net, rub any rocks that you used to anchor the net to the stream bottom and remove the rocks from the bottom. Firmly grab the bottom of the net so that your sample does not fall from the net, and then remove it from the water with a forward-scooping motion. The idea is to remove the net without allowing any insects to be washed under or off it.

Placing a white trash bag or white sheet under the net before separating the sample will catch any tiny organisms that may crawl through the net. Use a watering can or spray bottle to periodically water your net. The organisms will stop moving as the net dries out. Occasionally wetting the net will cause the organisms to move, making them easier to spot.

Watering the net is especially important on hot, dry days. Place the net on a flat, bright area, out of direct sunlight. Using tweezers or your fingers, separate all the organisms from the net and place them in your collecting container, which should be half full of water from the stream. Sort organisms into similar groups as you separate your sample. This will make your identification quicker when you are ready to record results. Plastic ice cube trays are helpful when sorting the catch. For example, put all organisms with legs in one section and all organisms with no legs in another section. Any organism that moves, even if it looks like a worm, is part of the sample. Look closely, since most aquatic macroinvertebrates are only a fraction of an inch long.

## **IDENTIFICATION**

Once organisms are collected through either the Rocky Bottom or Muddy Bottom Sampling methods, they are sorted and identified. You can use IWLA's "Field Guide to Aquatic Macroinvertebrates" or A Guide to Aquatic Insects and Crustaceans. IWLA's free Aqua Bugs app provides easy-to-follow instructions to help you identify your macroinvertebrates. Search for it in the Apple Store and Google Play Store.

Izaak Walton League macroinvertebrate guides provide a general overview of the macroinvertebrate types found across the United States. The composition of macroinvertebrate populations varies depending on local geography and geology. Try contacting your local environmental protection agency or universities for more information about local macroinvertebrates. Local experts might be able to share additional field guides that are specifically designed for your area.

Not all organisms in your stream are listed in the guides. For instance, macroinvertebrates such as whirligig beetles, water striders, and predaceous diving beetles are not included on the survey sheet. They are surface breathers and do not provide any indication of water quality.



When beginning your identification, ask yourself the following questions:

- How large is the organism?
- Is the body long and slender, round, or curved?
- Does the organism have any tails? How many?
- Does the organism have any antennae?
- Does the organism have legs? How many? Where?
- Is the body smooth and all one section, or is it segmented (two or more distinct sections)?
- Does the organism have any gills (fluffy or plate-like appendages)?
- Where are the gills located? Sides, back, underside, under its legs?
- Does it have pinching jaws like a beetle larvae?
- Are any legs or antennae missing because they were broken off in the net?
- What color is the organism?
- Does the organism swim underwater or remain on the surface?

When using the macroinvertebrate guides, read the descriptions for each organism. Sizes are provided for reference. However, if you catch a young macroinvertebrate that has just hatched and has not yet reached full size, it may be smaller than indicated in the guides. Specimens can be put into magnifying boxes to ease identification.

After identifying the organisms, record your results on the Biological Monitoring Data Form. Include information relating to habitat and physical parameters of the stream. Return the organisms to the stream after sampling is completed.

Tabulate your results to determine water quality using the instructions on the data form. Use letters to indicate the number of each type of organism (A=1–9, B=10–99, C=100 or more). Add the number of letters in a column and multiply by the index value at the bottom. Add the subtotal for each column to arrive at your final stream rating.

You will notice that the letter (A, B, or C) does not affect the final rating score of excellent, good, fair, or poor. This is because the survey is based primarily on diversity, not the number of individual organisms found. However, the letters are valuable because they document changes in populations over time. For example, your spring survey has only C's in the "pollution sensitive" column and only A's in the "pollution tolerant" category. In your summer survey, you find only A's in the sensitive range and C's in the tolerant range. Although your rating would remain the same, you might conclude that overall water quality was declining because populations of the tolerant organisms are increasing (A to C) while those in the sensitive category are decreasing (C to A). You should monitor for an entire year to get a clear picture of your stream health.

## **STREAM PROBLEMS AND THEIR EFFECTS ON STREAM ORGANISMS**

1. Physical Problems may include excessive sediment from erosion, street runoff, or discharge pipes. Sediment can create poor riffle characteristics, contribute to excessive flooding, reduce flow, change water temperature, and smother aquatic life. The result is usually a reduction in the number of macroinvertebrates in the study area.
2. Organic Pollution is from excessive human or livestock wastes or high nutrient enrichment from farm or yard runoff. The result is usually a reduction in the diversity of insects.
3. Toxic Pollution includes chemical pollutants such as chlorine, acids, metals, pesticides, and oil. The result is usually a reduction in the number of insects.

<b>Observation</b>	<b>Analysis</b>
<ul style="list-style-type: none"> <li>● High diversity, high numbers; many sensitive species such as stoneflies, caddisflies, and mayflies</li> </ul>	<ul style="list-style-type: none"> <li>● No problem; good water quality</li> </ul>
<ul style="list-style-type: none"> <li>● High diversity, low numbers</li> </ul>	<ul style="list-style-type: none"> <li>● Possibly due to poor habitat conditions</li> </ul>
<ul style="list-style-type: none"> <li>● Low diversity, high numbers</li> </ul>	<ul style="list-style-type: none"> <li>● Organic pollution (nutrient enrichment) or sedimentation; excessive algae growth from nutrient enrichment</li> </ul>
<ul style="list-style-type: none"> <li>● Low diversity, low numbers; or no bugs found but the stream appears clean</li> </ul>	<ul style="list-style-type: none"> <li>● Toxic pollution (e.g., chlorine, acids, heavy metals, oil, herbicides, insecticides)</li> </ul>

# SOS Macroinvertebrate Tally Sheet



**SAVE OUR STREAMS**

IZAAK WALTON LEAGUE OF AMERICA



## Biological Monitoring Data Form for Stream Monitors

(Modified for use by North Carolina Aquatic Data Hub)

Your Name: \_\_\_\_\_ Site ID: \_\_\_\_\_  
 Group Name: \_\_\_\_\_ Number of Participants: \_\_\_\_\_  
 Name of Stream: \_\_\_\_\_ GPS Coordinates: \_\_\_\_\_  
 City/State: \_\_\_\_\_ Survey Date: \_\_\_\_\_ Start Time: \_\_\_\_\_ End Time: \_\_\_\_\_  
 Description of Site Location: \_\_\_\_\_

### ROCKY BOTTOM SAMPLING

Before sampling, record the riffle composition on the back of this form. Using a kick-siense net, take one 60-second sample in a riffle area (40 seconds to rub rocks, 20 seconds to disturb the streambed). Ensure you sample the entire 3'x 3' area in front of the net. If you do not collect at least 100 macroinvertebrates in the first net, take a second sample in the same riffle. Please place a checkmark next to the number of samples collected.

\_\_\_\_\_ Sample 1      \_\_\_\_\_ Sample 2

### MUDDY BOTTOM SAMPLING

Record the total number scoops taken from each habitat type (20 scoops total) and provide details to best describe the specific habitat on the lines below.

Steep bank/vegetated margin \_\_\_\_\_  Woody debris with organic matter \_\_\_\_\_  
 Rock/gravel/sand substrate \_\_\_\_\_  Silty bottom with organic matter \_\_\_\_\_

### MACROINVERTEBRATE COUNT

Please consult biological monitoring instructions on how to conduct the macroinvertebrate count. Use the attached tally sheet to track numbers of each macroinvertebrate found. Once sampling and identification are complete, place a check mark next to each type of macroinvertebrate identified and list the total number found. Add up the number of checkmarks in each category (sensitive, less sensitive, tolerant) and multiply those numbers by the indicated index value.

Sensitive (Ex: <input checked="" type="checkbox"/> 10 Caddisflies)	Less Sensitive (Ex: <input checked="" type="checkbox"/> 2 Dobsonflies)	Tolerant (Ex: <input checked="" type="checkbox"/> 3 Leeches)
<input type="checkbox"/> _____ Caddisflies (except net spinners)	<input type="checkbox"/> _____ Dobsonflies <input type="checkbox"/> _____ Crayfish	<input type="checkbox"/> _____ Aquatic worms
<input type="checkbox"/> _____ Mayflies	<input type="checkbox"/> _____ Fishflies <input type="checkbox"/> _____ Scuds	<input type="checkbox"/> _____ Black flies
<input type="checkbox"/> _____ Stoneflies	<input type="checkbox"/> _____ Crane flies <input type="checkbox"/> _____ Aquatic sowbugs	<input type="checkbox"/> _____ Midge flies
<input type="checkbox"/> _____ Watersnipe flies	<input type="checkbox"/> _____ Damselflies	<input type="checkbox"/> _____ Leeches
<input type="checkbox"/> _____ Riffle beetles	<input type="checkbox"/> _____ Dragonflies <input type="checkbox"/> _____ Clams	<input type="checkbox"/> _____ Lunged snails
<input type="checkbox"/> _____ Water pennies	<input type="checkbox"/> _____ Alderflies <input type="checkbox"/> _____ Mussels	
<input type="checkbox"/> _____ Gilled snails	<input type="checkbox"/> _____ Common net spinning Caddisflies	
_____ # of check marks multiplied by 3 = _____	_____ # of check marks multiplied by 2 = _____	_____ # of check marks multiplied by 1 = _____
Now add the three totals from each column for your stream's index value. Total index value = _____		
Total number of macroinvertebrates in sample: _____		

Compare the final index value to the following ranges of numbers to determine the water quality of the stream sample site.

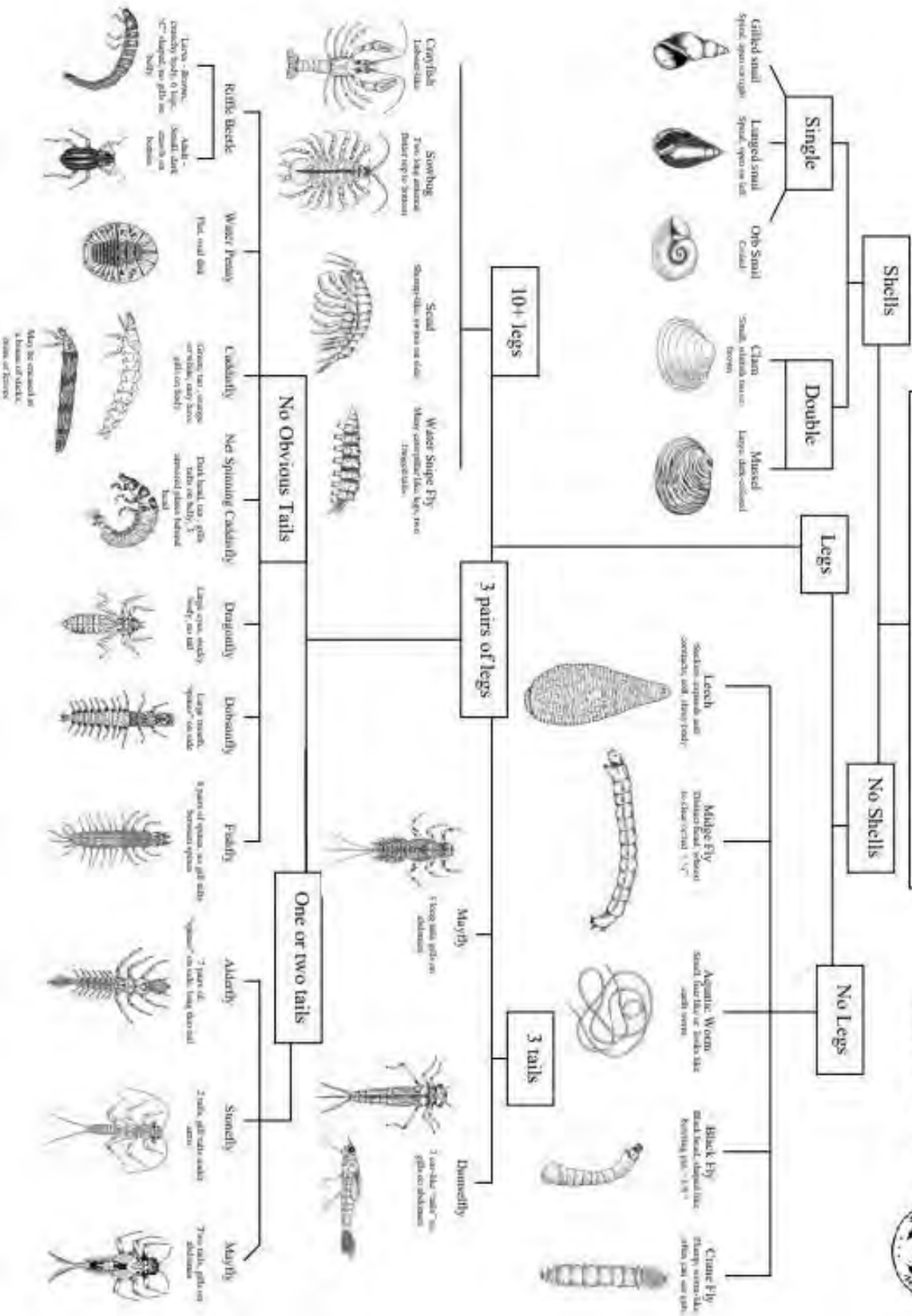
### WATER QUALITY RATING

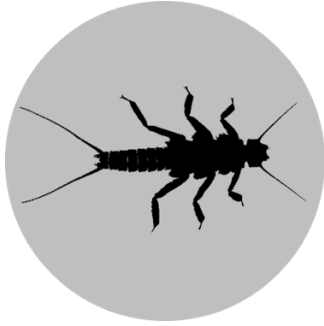
\_\_\_\_\_ Excellent (> 22)    \_\_\_\_\_ Good (17-22)    \_\_\_\_\_ Fair (11-16)    \_\_\_\_\_ Poor (< 11)

Share your stream monitoring data at [www.saveourstreams.net](http://www.saveourstreams.net).

# Key to Stream Macroinvertebrates

Izaak Walton League  
Save Our Streams





## SMIE Benthic macroinvertebrate assessment

**Tier 2**

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Developed by the Stream Monitoring Information Exchange (SMIE) of The Environmental Quality Institute ([www.eqilab.org](http://www.eqilab.org))

### Equipment provided by the organization:

Kick net (500 µm mesh or smaller)  
Collapsible table  
Boots/waders  
Deep, white, dishwashing bins (2)  
Forceps (4)  
Ice cube trays (4)  
Hand lenses (2)  
Clipboard with data and habitat sheets  
Pencils (3)  
Glass vials with alcohol for Quality Assurance samples (4)  
5-Gallon buckets (2-optional)  
Strainer for leaf-pack (optional - designed to fit over 5-gallon bucket for filtering biodiesel)  
D-frame or dip net with 1/32-inch mesh (optional)

### Personal equipment:

Laminated macroinvertebrate identification sheet  
Laminated protocol sheet  
Map/directions  
Watch

### Method: (requires 2-4 participants, including one certified Group Leader)

#### A. Site Selection and Timing

1. Choose wadeable streams greater than six feet across.
2. If you must sample near a bridge, go upstream or at least 50 feet downstream.
3. Adequate space must be available near the streambank for setting up the identification station (including folding table).
4. Choose sites in collaboration with the sponsor organization, based on sampling suitability and monitoring history.

5. Sampling is conducted in the spring and fall of each year.

B. Kick Net (Riffle) Sampling (2 people)

1. Riffles must be safely wadeable, preferably 10-30 inches deep, with a gravel to cobble bottom substrate. Representative riffles must have been covered in water through all seasons or else few to no macroinvertebrates may be found.
2. Place the kick net at the downstream end of the 15 ft<sup>2</sup> (5' long x 3' wide) section you wish to sample. Do not enter the sampling area until you are ready to kick.
3. Place the kick net along the bottom of the stream (weight with rocks to make sure), facing directly upstream into the current. Lean the net back but do not let water flow over the top of the net or you will lose organisms.
4. Kick and dislodge stones for one minute (use a stopwatch or timer).
5. As you remove the net from the water slowly, push forward and up (in a sweeping arc) to wash sampling material back into the net. If you lose the majority of your material, redo the procedure in another 15 ft<sup>2</sup> of the stream bottom.
6. Carry the net to the identification station (table), taking care to not lose organisms. Keep separate from leaf pack and visual samples.
7. Two people should pick invertebrates off the net, using forceps, for 20 minutes. Try to get at least 150 organisms off the net, and place them in ice-cube trays filled with stream water. If less than 150 are found, consider doing another full kick net sample (taken in a different location). There is no need to do three kick net samples. After picking for 20 minutes, time spent identifying and sorting organisms is unlimited.

C. D-Frame Net (Muddy bottom) Sampling (1 person) - OPTIONAL

1. If no riffles are present, the substrate is muddy or silty, and/or the stream flow is slow to stagnant, use a D-frame net (a.k.a. dip-net, sweep net) instead of the kicknet.
2. Target multiple habitats with this sampling technique, including the silty/muddy bottom, woody debris, rocks, submerged vegetation, and submerged roots along the streambank. Make sure that all habitats have been continuously submerged during all seasons. Approach the sample area from downstream and do not enter area until you are ready to start collecting.
3. Move the net in one-foot long sweeping or jabbing motions in upstream or bottom-to-surface directions along the targeted habitat. Make sure to dislodge specimens from solid substrates (wood, rocks) and scoop them from soft sediments and vegetation.
4. Perform 20 sweeps/jabs with the D-frame net from a variety of stream habitats. Move the mesh bottom of the net back and forth through the water (not allowing water to run over the top) to rinse fine silt from the sample.
5. Wash the contents of the net into a bucket or deep white bin containing water. This can be done multiple times until 20 sweeps/jabs are performed. Once all sweeps are collected, pour the entire sample through a net (e.g. kick net or approved strainer) for picking. Keep separate from leaf pack and visual samples.
6. Two people should pick invertebrates off the net, using forceps, for 20 minutes. Try to get at least 150 organisms off the net and place them in ice-cube trays filled with stream water. If less than 150 are found, consider doing another 10 sweeps/jabs (taken in different locations). There is no need to do additional collecting. After picking for 20 minutes, time spent identifying and sorting organisms is unlimited.

#### D. Leaf Pack Sampling (1 person)

1. Only collect leaf packs, which are submerged in flowing water. Look at the leaf packs carefully to make sure they are appropriate for sampling. You should see discoloration or physical decomposition of leaf pack, (especially 'lace-like' appearance where insects have been consuming leaf matter).
2. Take no longer than 3 minutes to make your leaf pack collections, or until you have an inch of leaf material in the bottom of the deep white pan. Return to the identification station. Keep the leaf pack sample separate from the kick net and visual samples.
3. Add stream water to pan and agitate the leaves before pouring water through the strainer or a corner of the kick net. Do this 2-3 times. One person should pick through the strainer and leaf debris for 10 minutes, placing the organisms in ice-cube trays filled with stream water.

#### E. Visual Inspection and Survey (1 person, preferably Group Leader)

1. Turn over rocks and logs in the stream. Examine roots along the streambanks. Pay careful attention since macroinvertebrates can be difficult to see. Stay in the stream and don't collect any insects that are not in the water.
2. Do not destroy habitats during your search and be careful to replace any objects you turn over during sampling.
3. Take 5 minutes to collect macroinvertebrates and take your sample to the identification station. Keep this sample separate from the kick net and leaf pack samples.

#### F. Habitat Survey

1. Answer the questions given on your data sheet that pertain to the characteristics of your sample site. If you are unsure of the meaning of any words or how to answer the questions, ask your Group Leader. This procedure should take about 10 minutes.

#### G. Sorting and Identification of Macroinvertebrates

1. Use the identification materials and the time provided to sort the insects and other organisms you have collected into rough groups (all mayflies, stoneflies, etc). Make sure you keep the three samples (kick net OR D-frame net, leaf pack, and visual) separate during sorting.
2. Use ice cube trays or other partitioned containers to put similar insects together. Group the sample into appropriate taxa before attempting to count or identify aquatic macroinvertebrates (counts and identifications do not have a time limit).
3. Look very closely at the contents of ice cube trays (smaller organisms get caught up in the legs or undersides of the larger organisms)
4. At the end of the time limits, identify all organisms, tally your results, and write on the data sheets provided. Consistent and legible data recording is important! Switching from numbers to tick marks may lead to errors (24+III = 135 or 27?).
5. Every season, Group Leaders will preserve all organisms from only one sample site for Quality Assurance. Place organisms in alcohol-filled vials before cleaning out the trays (one vial for each type of sample, plus one for any unidentified organisms). In a small corner of a data sheet, write in pencil the Group Leader name, sample site, date, and sample-type (kick net, D-frame net, leaf pack, or visual). Rip off this information and place it in the appropriate vial with the specimens.

6. Clean all equipment with stream water. Sampling gear, data sheets, and preserved specimens should be returned to the organization. Data analysis and site ratings are conducted by the sponsor organization.



### SMIE Biomonitoring Field ID Sheet

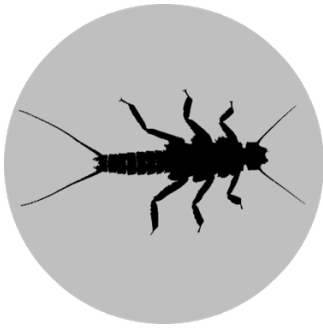
Stream: \_\_\_\_\_ Nearby Road: \_\_\_\_\_ County: \_\_\_\_\_ Date: \_\_\_\_\_

Group Leader: \_\_\_\_\_ Volunteers: \_\_\_\_\_ Weather: \_\_\_\_\_

		KICK NET	Total	LEAF PACK	Total	VISUAL	Total
		<b>STONEFLY</b>					
	1. Giant Shredder						
	2. Roach Shredder						
	3. Quick Crawling Predator						
	4. Fragile Derrivore						
		<b>MAYFLY</b>					
	5. Trinecaled Flattened Scraper						
	6. Two-tailed Flattened Scraper						
	7. Spiny Crawler						
	8. Round Headed Swimmer						
	9. Burrowing Mayfly						
	10. Spiny Turtle Mayfly						
	11. Filter Mayfly						
		<b>CADDISFLY</b>					
<b>FREE-LIVING</b>	12. Common Net Spinner						
	13. Striped Net Spinner						
	14. Finger Net Caddis						
<b>ORGANIC CASES</b>	15. Small Head Caddis						
	16. Stick Bait Caddis						
	17. Square Log Cabin						
<b>MINERAL CASES</b>	18. Sand and Stick						
	19. Vegetative Case						
	20. Gravel Coffin Case						
	21. Sand Snail Case						
	22. Sand/ Mineral Case						
		<b>BEETLES</b>					
	23. Water Penny						
	24. Predator Beetle						
	25. Acut Rifle Beetle						
	26. Larval Rifle Beetle						
		<b>MEGALOPTERAN</b>					
	27. Hellgramite						
	28. Fishfly						
	29. Alderfly						
		<b>DIPTERAN</b>					
	30. Watergnat						
	31. Water-worm						
	32. Fat-headed Cranefly						
	33. Chironomid Midge						
	34. Red Midge						
	35. Blackfly						
	36. Oligochaete						
	37. Leech						
		<b>BIVALVES</b>					
	38. Mussels and Clams						
		<b>SNAILS</b>					
	39. Coiled Left Face Snail						
	40. Coiled Right Face Snail						
	41. Rounded Right Face Snail						
		<b>CRUSTACEANS</b>					
	42. Crayfish						
	43. Sowbug (Isopod)						
	44. Scud (Amphipod)						
		<b>ODONATES</b>					
	45. Damselfly						
	46. Dragonfly						

Total Kicknet #:	Total Leafpack #:
------------------	-------------------

Please write NOTES on the back (e.g. if you collected more than one sample, if you preserved the samples, if you threw out some specimens in the preserved sample).



## BAB Benthic macroinvertebrate assessment



The purpose of this section is to provide a high level summary of the standard operating procedures (SOPs) of the Biological Assessment Branch (BAB) of the North Carolina Division of Water Resources (DWR) for the collection and analysis of freshwater benthic macroinvertebrate data that may result in bioclassification (ratings).

This methodology requires significant benthos experience. Under this methodology, most organisms may be identified using a dissecting microscope, but Oligochaeta, Chironomidae, and some mayfly structures must be mounted on glass slides and identified with a compound microscope. Insects, including Chironomidae, are identified to genus or species; other groups are often identified to genus as well, but are sometimes left at higher taxonomic levels. Following identification, samples—both the portion preserved in ethyl alcohol and any microscope slides—are labeled, properly stored, and retained indefinitely. Quality assurance by a BAB-approved taxonomist is required for at least 10% of the samples collected.

Consistency in data collection and analysis is the cornerstone for evaluating biological integrity. The procedures followed by DWR are a synthesis of widely used methodologies developed from the experience of personnel within the branch. These methods have been shown to provide repeatable and useable data for water quality evaluations. Benthic macroinvertebrates, especially aquatic insects, are associated with the substrates of streams, rivers, and lakes. The BAB uses aquatic macroinvertebrate biological surveys as one type of indicator of biological integrity in streams and rivers.

To view the entire SOP please go to <https://deq.nc.gov> and search for “Benthic Macroinvertebrate Assessment Data,” or click through the following sequence of pages, to find the page where the current SOP is linked:

[About > Divisions > Water Resources > Water Sciences > Biological Assessment Branch > DWR Benthos Data](#)

If you’d like to explore this option further, you may contact:

Eric Fleek, Biologist Supervisor	<a href="mailto:Eric.Fleek@ncdenr.gov">Eric.Fleek@ncdenr.gov</a>	919.743.8469
Lauren Housley, Benthic Biologist	<a href="mailto:Lauren.Housley@ncdenr.gov">Lauren.Housley@ncdenr.gov</a>	919.743.8470

## Appendix 1. Temperature Conversion Table

### Degrees Centigrade to Degrees Fahrenheit

$$C^{\circ} = (F^{\circ} - 32) \times 5/9$$

<u>°C</u> - <u>°F</u>	<u>°C</u> - <u>°F</u>		
0.0	32.0	20.0	68.0
0.5	32.9	20.5	68.9
1.0	33.8	21.0	69.8
1.5	34.7	21.5	70.7
2.0	35.6	22.0	71.6
2.5	36.5	22.5	72.5
3.0	37.4	23.0	73.4
3.5	38.3	23.5	74.3
4.0	39.2	24.0	75.2
4.5	40.1	24.5	76.6
5.0	41.0	25.0	77.0
5.5	41.9	25.5	77.9
6.0	42.8	26.0	78.8
6.5	43.7	26.5	79.7
7.0	44.6	27.0	80.6
7.5	45.5	27.5	81.5
8.0	46.4	28.0	82.4
8.5	47.3	28.5	83.3
9.0	48.2	29.0	84.3
9.5	49.1	29.5	85.1
10.0	50.0	30.0	86.0
10.5	50.9	30.5	86.9
11.0	51.8	31.0	87.8
11.5	52.7	31.5	88.7
12.0	53.6	32.0	89.6
12.5	54.5	32.5	90.5
13.0	55.4	33.0	91.4
13.5	56.3	33.5	92.3
14.0	57.2	34.0	93.2
14.5	58.1	34.5	94.1
15.0	59.0	35.0	95.0
15.5	59.9	35.5	95.9
16.0	60.8	36.0	96.8
16.5	61.7	36.5	97.7
17.0	62.6	37.0	98.6
17.5	63.5	37.5	99.5
18.0	64.4	38.0	100.4
18.5	65.3	38.5	101.3
19.0	66.2	39.0	102.2
19.5	67.1	39.5	103.1

## Appendix 2. Equipment Information

### pH Testing

#### I. Colorimetric Kit: LaMotte Precision pH Kit

Clarkson Lab: Catalog# 5858-01 Precision pH Kit, OS2 pH 3.0-10.5 @ \$44.50

Note: Refill for the Wide Range Indicator solution: Clarkson Lab: #Lam2218-6 @\$7.02

#### II. Hach Pocket Pro+ Tester - measures pH and temperature

Hach: Item#9531000 @ \$78

Note: only contains small pocket of pH 7 buffer; would also need pH 4.0 and pH 10.0 buffers

#### III. pH test strips: pH 0 -> pH 14: Fisher Scientific: #13-640-508 @ \$17.14 - 100 test strips

### Dissolved Oxygen

#### I. Colorimetric Kit

Cole Parmer # EW-0554040 @ \$72; enough for 30 tests

#### II. Extech Dissolved Oxygen Meter

Cole Parmer #EW-53026-20 @ \$249.99

### Conductivity

Oaktron WD-35462-11 EcoTestr CTS - measures conductivity, salinity and TDS

Global Test Supply @54.20

Conductivity Standard: 20 mS/cm: Fisher Scientific: Ricca# 224716, 500 ml bottle@ \$35.75

### E.coli/coliform

Micrology Lab: CWK10 Coliscan Water Monitoring Kit @ \$35.15

Incubators: Agri-Supply Item# 21113 @ \$56.99; 1-800-345-0169

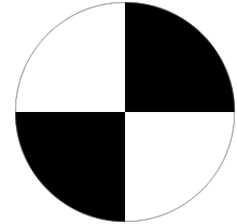
### Appendix 3. Example Equipment Resolution and Accuracy

Parameter	Equipment or Test Kit	Reported Resolution	Reported Accuracy
	Colorimetric strips; LaMotte Wide range pH kit	None	None
pH	Hach Pocket Pro + Testr	0.1 pH	±0.5 pH
	YSI ProPlus	0.01 pH	±0.2 pH
	CHEMetrics Ampoules;	None	None
Dissolved Oxygen	Extech DO pen meter	0.1 mg/L	±0.5 mg/L
	YSI ProPlus	0.01 mg/L	± 0.2 mg/L
Temperature	Fisher scientific traceable lollipop thermometer	0.1 °C	±1°C
	YSI ProPlus	0.1°C	±0.2°C
Chloride	YSI ProPlus, In-situ smarTROLL, Hydrolab MS5	Chloride: 0.01 mg/L	Chloride: 5 mg/L
Salinity	Oakton EcoTestR CTS, YSI ProPlus, In-situ smarTROLL, Hydrolab MS5	0.1 ppt	2% FS
	YSI ProPlus, In-situ smarTROLL, Hydrolab MS5	0.1 ppt	0.01 ppt
Total Dissolved Solids	Oakton EcoTestR CTS, YSI ProPlus, In-situ smarTROLL, Hydrolab MS5	0.1 ppt	2% FS
	YSI ProPlus, In-situ smarTROLL, Hydrolab MS5	0.1 ppt	0.01 ppt
Chlorophyll A	YSI EXO2, grab sample - lab analysis		Sonde: 0.01 µg/L Chl;
	YSI EXO2, grab sample - lab analysis		Sonde: 0.01 µg/L Chl;
	DIY tube; Lawrence Enterprises TTG-120CM	None	None
Turbidity	Sper Scientific Turbidity Meter	Turbidity: 0.01 NTU	±5% F.S.
	YSI EXO2	Turbidity: 0.01 NTU	Turbidity: 0.3 NTU

## Appendix 4. Turbidity tube construction and CM to NTU Table

### Constructing a Turbidity Tube

1. Obtain a Fluorescent Light Tube Cover at any hardware store.
2. Obtain a 1-¼ inch white PVC cap with flat bottom.
3. Draw a Secchi disc (see figure) on the inside of the PVC cap using a black magic marker.
4. Hold a tape measure along the side of the tube. Mark the distance along the tube in centimeter increments, using the bottom of the tube as the starting point.
5. Glue the PVC cap to the bottom of the Turbidity Tube.
6. If desired, mark the 85.4 cm mark on the tube, indicating the maximum range of clarity that can be measured by this method.



Centimeters	NTU
6.7	240
7.3*	200*
8.9	150
11.5	100
17.9	50
20.4	40
25.5	30
33.1	21
35.6	19
38.2	17
40.7	15
43.3	14
45.8	13
48.3	12
50.9	11
53.4	10
85.4*	5*

**Table. Centimeter to NTU conversions**

## Appendix 4. North Carolina Water Quality Classifications and Standards

### Background

Many NC waters were initially assessed in the 1970s. These initial assessments were used to determine uses for the different waters. Classifications were then assigned to waters to define uses to be protected. Finally, standards (associated with classifications) were developed to protect uses associated with classifications.

### What are Surface Water Classifications?

Surface Water Classifications are designations applied to surface water bodies, such as streams, rivers and lakes, which define the best uses to be protected within these waters (for example swimming, fishing, drinking water supply) and carry with them an associated set of **water quality standards to protect those uses**. Surface water classifications are one tool that state and federal agencies use to manage and protect all streams, rivers, lakes, and other surface waters in North Carolina. Classifications and their associated protection rules may be designed to protect water quality, fish and wildlife, or other special characteristics. **Each classification has associated standards that are used to determine if the designated uses are being protected.**

### What Are Water Quality Standards?

Water quality standards are state regulations or rules that serve to protect the lakes, rivers, streams, and other surface waters of the state from the deleterious effects of pollution. Surface waters are protected based on their designated "best uses" as defined in the surface water classifications established in Title 15A of the [North Carolina Administrative Code](#) (NCAC) subchapter 02B. For more information on surface water classifications, please see the [Surface Water Classifications](#) webpage.

Standards for parameters included in this manual:

Parameter	Standard	Comments
Conductivity (specific conductance)	No NC standard	See text below table
Dissolved Oxygen (mg/L)	$\geq 5.0$ , $\geq 4.0$ (I), $\geq 6.0$ (T)	I-Instantaneous, T=trout, may be exceptions for swamp waters, etc.

Fecal Coliform	(see narrative)	
pH	6.0-9.0, 6.8-8.5 (SC)	SC=saltwater
Temperature	(see narrative)	
Turbidity (NTU)	<=50, <=25 (RL, SC), <=10 (trout)	RL = reservoirs, lakes, etc.

Conductivity is a measure of the ability of water to pass an electrical current. Conductivity in water is affected by the presence of inorganic dissolved solids such as chloride, nitrate, sulfate, and phosphate anions (ions that carry a negative charge) or sodium, magnesium, calcium, iron, and aluminum cations (ions that carry a positive charge). Organic compounds like oil, phenol, alcohol, and sugar do not conduct electrical current very well and therefore have a low conductivity when in water. Conductivity is also affected by temperature: the warmer the water, the higher the conductivity. For this reason, conductivity is reported as conductivity at 25 degrees Celsius (25 C). The basic unit of measurement of conductivity is the mho or siemens (USEPA). Dissolved oxygen is the amount of oxygen present in the water. Water bodies receive oxygen from the atmosphere and from aquatic plants. Running water, such as that of a swift moving stream, dissolves more oxygen than the still water of a pond or lake (USEPA).

Fecal coliform bacteria are a subgroup of Total coliform bacteria (a collection of relatively harmless microorganisms that live in large numbers in the intestines of man and warm- and cold-blooded animals). They aid in the digestion of food. Fecal coliform bacteria (most common member being *Escherichia coli*) may be separated from the total coliform group by their ability to grow at elevated temperatures and are associated only with the fecal material of warm-blooded animals. Fecal coliform by themselves are usually not pathogenic; they are indicator organisms, which means they may indicate the presence of other pathogenic bacteria. Pathogens are typically present in such small amounts it is impractical monitor them directly (Water Research Center).

**Fecal coliform: shall not exceed a geometric mean of 200/100ml (MF count) based upon at least five consecutive samples examined during any 30 day period, nor exceed 400/100ml in more than 20 percent of the samples examined during such period. Violations of the fecal coliform standard are expected during rainfall events and, in some cases, this violation is expected to be caused by uncontrollable nonpoint source pollution. All coliform concentrations shall be analyzed using the membrane filter technique, unless high turbidity or other adverse conditions necessitate the tube dilution method. In case of controversy over results, the MPN 5-tube dilution technique shall be used as the reference method.**

**pH is a measure of how acidic/basic water is. The range goes from 0 - 14, with 7 being neutral. pHs of less than 7 indicate acidity, whereas a pH of greater than 7 indicates a base (USGS)**

**Temperature: not to exceed 2.8 degrees C (5.04 degrees F) above the natural water temperature, and in no case to exceed 29 degrees C (84.2 degrees F) for mountain and upper piedmont waters and 32 degrees C (89.6 degrees F) for lower piedmont and coastal plain Waters; the temperature for trout waters shall not be increased by more than 0.5 degrees C (0.9 degrees F) due to the discharge of heated liquids, but in no case to exceed 20 degrees C (68 degrees F);**

**Turbidity is the measure of relative clarity of a liquid. It is an optical characteristic of water and is an expression of the amount of light that is scattered by material in the water when a light is shined through the water sample. The higher the intensity of scattered light, the higher the**



**turbidity. Material that causes water to be turbid include clay, silt, finely divided inorganic and organic matter, algae, soluble colored organic compounds, and plankton and other microscopic organisms (USGS).**