SOUTH CAROLINA ADOPT-A-STREAM VOLUNTEER FRESHWATER MONITORING HANDBOOK — EDITION 3 —





OCTOBER 2024

SOUTH CAROLINA ADOPT-A-STREAM CONTACTS

If there is evidence of criminal activity, leave the area immediately and call local law enforcement.

If there is evidence of dangerous pollution discharges, fish kills, or public health hazards, report it to SCDES's Emergency Hotline at 1-888-481-0125.

If your bacteria result is higher than 1000 CFU/100 mL, please go back and resample as soon as possible. If your second sample result is still greater than 1000 cfu/100 mL, a SC Adopt-a-Stream program coordinator may contact you with more information. Both sampling events should be entered as "50" for each plate value that is considered "Too Numerous To Count" (TNTC) so that an alert is sent to program staff. In addition, enter "TNTC" in the comments box. The third photo option in the database should be used to photo document any perceived threat or pollution source observed during your sampling event.

Please remember to take detailed notes on the following:

- Exact location (address or GPS coordinates)
- The nature of the issue
- Date and time the issue occurred or started
- A picture of the site

Water quality parameters are sensitive to weather conditions, temperature, and rainfall. SC Adopt-a-Stream data are collected by trained citizen water quality monitoring volunteers. It is baseline, screening data for educational purposes, and is not used in any regulatory procedures.

SC Adopt-A-Stream Contacts

Contact SC Adopt-a-Stream staff by emailing scaas@des.sc.gov or by calling 803-898-4168. Answers to most frequently asked questions can be found at <u>www.scadoptastream.org</u>.

For questions about monitoring techniques, contact your SC AAS Trainer.

ACKNOWLEDGEMENTS

The South Carolina Adopt-a-Stream State Team would like to acknowledge and thank the many great programs that helped shape this SC Adopt-a-Stream Volunteer Freshwater Monitoring Handbook, now in its third edition. This training resource and backbone of our SC AAS program has immensely benefited from these fellow leaders in community science and their volunteers who have tested these materials over the past few decades. Specifically, we thank:

Georgia Adopt-A-Stream led by Georgia Environmental Protection Division IdaH2O Water Stewards led by University of Idaho Extension Service Watershed Watch led by University of Rhode Island Water Action Volunteers led by University of Wisconsin and Wisconsin Department of Natural Resources

The momentum to develop consistent SC Adopt-a-Stream tools and resources to better serve community scientists interested in healthy waterways was driven by the dedication of many organizations and individuals. For this, we thank:

Anderson County Anderson University Clemson Extension Service Clemson University The Enggasser Family Greenville Technical College Pickens County Richland County SC Department of Environmental Services SCDES Aquatic Science Programs Southern Wesleyan University Upstate Forever USC – Spartanburg Watershed Ecology Center

Special thanks to SCDES's Bureau of Water for their editorial contributions: James B. Glover, Ph.D., Manager of the Aquatic Biology Section (retired) David Chestnut, Senior Scientist, Surface Water Monitoring David Eargle, Aquatic Biologist Justin Lewandowski, Aquatic Biologist Lexi Thomason, SC Adopt-a-Stream Program Coordinator

In addition, special thanks for the insightful edits and suggestions from past and present SC Adopt-a-Stream Trainers, Kaleigh Sims, Beth Button, and Renee Gracon.

South Carolina Department of Environmental Services and the Clemson University Center for Watershed Excellence are proud to offer you this training opportunity and series of science-based resources to understand and better protect our shared, most precious natural resource.

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TO PROMOTE AND PROTECT SOUTH CAROLINA'S WATERWAYS THROUGH CERTIFIED VOLUNTEER WATER QUALITY AND ECOSYSTEM MONITORING. -SC ADOPT-A-STREAM MISSION STATEMENT

1.1 INTRODUCTION

Welcome to the South Carolina Adopt-a-Stream family. You are joining a team of like-minded individuals working to build awareness of watershed health. Volunteer monitoring contributes to the improved management and protection of South Carolina's waterways and ecosystems.

South Carolina is blessed with mountain streams, wetlands, black waters and slow-moving streams, estuaries and beaches, and the largest intact expanse of old growth bottomland hardwood forest remaining in the southeastern United States at Congaree National Park.

SC Adopt-a-Stream (SC AAS) provides the opportunity for those interested in the protection and improved management of South Carolina's waterways to be directly involved in their monitoring and reporting. Volunteer monitors provide vital baseline data that complements local and state data used to determine the health of our waterways. In sharing this information about stream conditions, volunteers, local communities, educators, and local government agencies can partner to protect and restore our waters. SC AAS trained volunteers have the potential to increase awareness within their own community of the relationship between pollution, watershed management, land use changes, and the personal responsibility of each individual within the watershed to be a better steward.

SC AAS volunteers can become certified to monitor freshwater streams, tidal saltwaters, macroinvertebrates (aquatic insects), or freshwater lakes. As a SC AAS Volunteer Freshwater Monitor, you will be trained in Stream Habitat Assessments and certified in Physical, Chemical, and Bacterial Monitoring. SC AAS seeks to create a network of information and resource sharing, helping connect volunteers with equipment and information, and providing a secure, mobile-friendly database to share water quality monitoring data. This handbook will serve as your reference for monitoring, as well as a resource for more information on why these monitoring data are meaningful to water resource management. Words in bold are found in Appendix 2, the Glossary.

PROGRAM INTRODUCTION

QUICK FACT

As volunteers, we take an Active approach to

collecting Data while enjoying the Outdoors in

order to help Protect our waters Together.

PROGRAM LEADERSHIP AND FUNDING

SC AAS is led by the SC Department of Environmental Services (SCDES) in partnership with many local organizations. Your Adopt-a-Stream Program Coordinators with SCDES keep the program running and growing while Clemson University manages the public database and online recertification. Other partners

serve as trainers, kit loan locations, and program supporters. Initial funding was provided by SCDES, Clemson University Public Service & Agriculture, and US Environmental Protection Agency (EPA) Region 4.

What does it mean to ADOPT?

The SC Adopt-a-Stream program seeks to:

- 1. Increase citizens' knowledge of water issues within their watershed.
- 2. Provide information about waterways and potential sources of pollution through our public database.
- 3. Lead to changes in behavior that begin with the responsibility of monitoring a waterway and ecosystem health.
- 4. Provide opportunities for watershed stewardship in action.



1.2 LEVELS OF INVOLVEMENT

Levels of involvement have been developed for several purposes and meet different training requirements.

- 1. Certified Volunteer Trained steward who is certified to collect data according to the procedures outlined by the SC AAS program. Volunteers attend a workshop and pass a written test and must recertify annually to continue sharing data in the SC AAS Database.
- 2. Certified Trainer Part of a network of mentoring and peer training, which increases partnerships with agencies, conservation organizations, and communities across the state. Trainers host at least two workshops per year, serve as a local resource for volunteers, and continue regular stream monitoring. Trainers renew their certification annually.
- 3. SC AAS "Hub" A coordinating entity that utilizes SC AAS in their programming and provides additional resources and mentoring to local volunteer monitoring groups. This may include the lending of kits, assisting with data interpretation, regularly hosting workshops, and more. Visit scadoptastream.org to learn more!

1.3 QUALITY OF PROGRAM AND DATA INTEGRITY

SC AAS is developed for educational purposes, first and foremost, and to provide baseline water quality data and additional watershed insights for resource managers at the local and state level. The standardization of trainings and resources, sampling protocols, and duplicate samples ensure consistency and integrity of the data collected. In freshwater stream systems, SC AAS trains and collects data under the following protocols and assessment, all contained within this Freshwater Monitoring Handbook:

- 1. Physical/Chemical Core measurements record dissolved oxygen (DO), air and water temperature, pH, and transparency.
- 2. Bacteria Focuses on Escherichia coli (E. coli), a subgroup of fecal coliform bacteria.
- 3. Stream Habitat Assessment Records and scores stream habitat qualities, such as streambank stability, presence of vegetation, and presence of riffles and pools.

The Macroinvertebrate Monitoring protocol for freshwater streams helps quantify the abundance and diversity of aquatic life and appraise the stream's health. More on this type of monitoring can be found at our website, <u>www.scadoptastream.org</u>.

A Quality Assurance Project Plan (QAPP) ensures consistency of data collection, trainings, handbook, and program policies and procedures. An updated QAPP was approved by US Environmental Protection Agency (EPA) Region 4 and SCDES in 2022 and is available on the SC AAS website, <u>www.scadoptastream.org</u>. It will be regularly reviewed by collaborators and can be modified and resubmitted as this SC AAS program evolves and grows. The QAPP is an important reference document that guides SC AAS policies and operations.

SC Adopt-A-Stream volunteers collect **baseline**, non-regulatory data on the health of the state's waterways. Rarely, certain water quality conditions or site observations may warrant a follow-up visit by local or state agencies to collect further information; however, the program is not intended to be used to target landowners or individuals. We as a SC AAS community are neighbors within our watersheds, encouraging practices and knowledge gain that protect what we value about our natural world.

USE OF DATA

While the screening level data generated by the program method does not meet the rigorous data quality requirements for SCDES regulatory decisions, it provides many benefits. Data collected by SC AAS

Volunteer Monitors is used to establish baseline conditions for determining stream health based on chemical, physical, biological, and habitat parameters. This data is useful in screening waterbodies for water quality problems, and in assessing the overall health of a watershed. The data may also be used to:

- Identify waterbodies in need of more detailed monitoring;
- Identify specific areas within a watershed in need of water quality improvement;
- Assist local watershed councils and partners in making environmental management decisions in their local and regional watersheds;
- Enlist community involvement in their local watershed;
- Prioritize areas in a watershed for Best Management Practices (BMPs);
- Assess BMP/remediation project performance;
- Identify potential pollution sources; and
- Provide educational and involvement opportunities for all interested in learning more about the health of their local waterways.

1.4 IDENTIFYING YOUR MONITORING OBJECTIVES

Volunteers come to the program with different goals, and these goals may direct the type(s) of monitoring you choose to conduct. Keep in mind that monitoring a waterway to establish baseline information is a long-term commitment. In very few cases is a water quality concern solved with just a few data points. Let's consider why you want to become a SC AAS Volunteer Monitor to help determine which type of monitoring is most suitable for your objectives.

WHY ARE YOU A MEMBER?

You care about the health of your watershed and want to contribute meaningful data toward the understanding and protection of local streams.

For a large-scale watershed study, you may choose to monitor more than one site, such as several feeder tributaries. You may not necessarily need many protocols at first. The Stream Habitat Assessment, which takes less than 20 minutes to perform, can be used to quickly record changing conditions. From this broader data gathering perspective, the volunteer can view trends in stream corridor health, identify potential problematic inputs, then further narrow down a more specific protocol for those waterways. This more widespread data collection can inform neighborhoods and may lead to the development of watershed organizations, building towards more resources and a motivated public watching and monitoring their local waterways.

You are concerned that growth and development may be impacting waterways with increasing stormwater volumes and velocities.

The Stream Habitat Assessment is requested only once per year, but it can be used more frequently (seasonally, for instance) to track streambank stability, sediment deposition, riparian changes, and parameters affected by extreme fluctuations in flow.

Discharges of high-temperature water can act as a pollutant by increasing the stream temperature, lowering dissolved oxygen, and impacting organisms. Temperature can be monitored using the Freshwater Stream Monitoring Protocol.



You care about the safety of swimming and recreation waters near your backyard. South Carolina offers innumerable recreation possibilities on the water, which are directly linked to our heritage, culture, economy, and quality of life. Fecal-related bacteria can threaten these uses and public health, making it the most critical element for monitoring. When recording site observations, one should be diligent in recording changes in flow, stream height, and stream width, as changes in available water are also critical considerations in the recreational use of a waterway.

You often smell or observe irregularities in stream conditions and feel that these instances are difficult to record, leaving you unsure of what to report and when to report concerns. It is important to report suspicions or observations when it comes to illegal discharges, spills, and wastewater overflows. Use SCDES's "Report It!" form located at https://des.sc.gov/about-scdes/contact-us/report-environmental-concern. Your report can expedite a response and action to protect the stream and its inhabitants.

The Freshwater Stream Monitoring Protocol includes temperature, dissolved oxygen, pH, transparency, and bacteria. These recordings organize observations and suspicions into quantitative means of communicating a potential problem. If observations include the smell of sewer waste, or you have observed black water or staining coming from pipes discharging to the waterway, bacterial monitoring may be the best means to identify an issue.

The SC AAS Database is built to send alerts to the SC AAS State Team when odor or color is recorded as "Other" or *E. coli* is 1000 cfu/100 ml or greater. In addition, South Carolina cities and counties can request these automated alerts for sampling sites in their jurisdiction. Therefore, your ability to report all conditions recorded at the stream is streamlined by the database.

You are an avid fisherman and are concerned that conditions in the stream and changing land uses may stress fish and their habitat, and lead to decline.

Decline of habitat suitability typically happens over a period of time. Macroinvertebrates, who build their communities in the stream, are indicators of improving or declining stream conditions. Macroinvertebrate Monitoring (a separate certification workshop) includes the quantitative count of organisms, identifying which are pollution sensitive or pollution tolerant, and qualitatively describes stream health based on its residents. The Macroinvertebrate Protocol also records observations about the stream, including habitat type and availability.

The Stream Habitat Assessment includes more specific features such as channel stability, sinuosity, and aquatic vegetation, which can further describe changes in habitat suitability.

1.5 SC AAS VOLUNTEER CODE OF ETHICS

SC AAS Volunteer Monitors and Trainers represent this program with integrity by doing the following:

- Following the SC AAS monitoring methods;
- Fully and accurately documenting observations;
- Reporting data directly to the SC AAS database; and
- Accurately and respectfully discussing the program with others.

It is also expected that certified volunteer monitors foster positive relationships with landowners, inviting all audiences to be stewards of our shared water resources, by:

- Seeking written permission to access private property, if necessary, to monitor a stretch of waterway. A landowner permission form can be downloaded from the "Resources and Materials" page of our website;
- Always being clear and truthful in identifying themselves as a certified volunteer monitor of SC AAS, as well as the intended use of monitoring results;
- Making every effort to contact the landowner in advance with exact date(s) of sampling or an agreed upon monitoring schedule;
- Never misusing or harming private property;
- Taking extra precautions for everyone's personal safety when on private property; and
- Making good faith efforts to regularly share results of monitoring with landowners.

1.6 SAFETY IS OUR FIRST PRIORITY

Practicing safe sampling begins during your monitoring site selection and underscores all field activities.

- Site selection includes the consideration of many factors distance from vehicle, access, steepness of slope, history of flash flooding, and general site safety.
- Always use the "buddy system" and sample with your group or another volunteer monitor.
- Always let someone else know that you are going out into the field and approximately how long sampling should take.
- Use caution when entering the stream, ensuring that you can safely exit, the current is not too strong, and the bottom will safely support you while standing and walking. Stop monitoring if a storm occurs while on-site.
- As a general rule of thumb, do not access water that is above your knees.
- Monitor during the day.
- Wear proper attire closed toed shoes, boots or waders, and enough clothing to protect your body from brush, insects, and UV rays.
- Use gloves and eye protection when performing chemical and bacterial monitoring protocols,
- Be cognizant of signs of heat stroke and hypothermia and how to treat these conditions. Stay hydrated.
- Always be aware of your surroundings, including traffic, livestock, poisonous plants, stinging insects, loose rock, and other people.
- Use soap and water or hand sanitizer at the completion of monitoring.
- If you believe that your monitoring location has experienced significant or harmful contamination, do not sample. Please discuss these concerns with SC AAS State Team members.
- STOP monitoring if you witness any illegal or suspicious discharge entering the waterway.

PROGRAM INTRODUCTION

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2.1 INTRODUCTION

A **watershed** is an area of land that drains all the streams and rainfall to a common outlet such as the outflow of a reservoir, mouth of a bay, or any point along a stream channel. The word "watershed" is sometimes used interchangeably with "river basin" or "catchment." Ridges and hills that separate two watersheds are called drainage divides. Rain and runoff from these divides drains to the streams in the watershed, as shown in the figure on the next page. Larger watersheds contain many smaller watersheds with different outflow points. Understanding the boundaries of your watershed and how it is affected

by the larger watershed provides insights on how land use, permitting, and other occurrences in the watershed may result in changes in your data and observations at your site(s).

There are eight major watersheds, or river basins, in South Carolina: the Savannah, Saluda, Broad, Catawba, Pee Dee, Edisto, Santee, and Salkehatchie.



Direction of water flow in a watershed (Source: Center for Watershed Protection)



Major River Basins of South Carolina (Source: SC Department of Natural Resources)

Each watershed is identified by a unique **Hydrologic Unit Code**, or "**HUC**." HUCs are useful for identifying which watershed you want to consider as you plan to monitor, as watersheds are not contained by city, county, or even state boundaries. Within the HUC system, the United States is divided and subdivided into successively smaller watersheds. As the watershed area gets smaller, the unique HUC numbers get longer. The SC AAS

Database organizes sites by 8-digit HUC.

Let's look at this example from the Broad River Watershed:

The Broad River Watershed, at greater than 2.5 million acres in size, is identified as 030501, a 6-digit HUC. It is further subdivided into 8-digit HUCs:

- the Enoree River Watershed, 03050108;
- the Tyger River Watershed, 03050107;
- the Upper Broad River Watershed, 03050105; and
- the Lower Broad River Watershed, 03050106.



Sub-watersheds (8-digit and 10-digit HUCs) within the Broad River Watershed.

WATERSHEDS & WATER

The Ecoregions of South Carolina

An **ecoregion** is a large unit of land or water containing a geographically distinct assemblage of species, natural communities, and environmental conditions. This includes geology, landforms, soils, vegetation,

climate, land use, wildlife, and hydrology. South Carolina is divided into five (Level 3) ecoregions, which will affect the stream habitats and parameters you are monitoring. For example, in the Blue Ridge and Piedmont ecoregions, you can encounter rocky and sandy streams. In



Level 3 Ecoregions of South Carolina

the Southeastern and Middle Atlantic Coastal Plains, you can encounter black water streams with naturally lower pH and dissolved oxygen levels. Tidal saltwaters are found along the Southern Coastal Plain.

2.2 DETERMINING THE HEALTH OF FRESHWATER SYSTEMS

The health and vitality of South Carolina's waterways are the cornerstone to all things South Carolina. Our waterways power our industries and homes, with power generation utilizing the greatest volumes of water in our permitting system. Our state's largest industry, agriculture, uses surface and groundwater to put food on our table and feed the growing local food movement. The state's residents and tourists paddle, swim, fish, and boat from our mountain streams to the coast. Ample rainfall, groundwater replenishment, and clean water help maintain a healthy South Carolina economy.

In the past two centuries, human activities have had a significant effect on South Carolina's water quality. Point and nonpoint source pollution contribute to water quality problems. **Point sources**, single identifiable sources of pollution, have been the focus of regulatory oversight for decades. Attention to point source problems has resulted in significant improvement in water quality. **Nonpoint source** pollution, unlike pollution from industrial and sewage treatment plants, comes from many diffuse sources. Nonpoint source pollution is caused by rainfall or snowmelt moving over and through the ground. As the runoff moves, it picks up and carries away natural and human-made pollutants, finally depositing them into lakes, rivers, wetlands, coastal waters, and ground waters. Nonpoint sources are now a substantial detriment to achieving designated uses for South Carolina's streams and rivers. SC AAS volunteers can play an important role in tracking and monitoring water quality and sharing information about local water resources with their communities.

Many "pollutants" in a stream are naturally occurring and are only considered a pollutant when their presence is impairing a waterway's ability to maintain life or are a potential threat to users. Therefore, assigning a numeric measure allows a resource manager to judge whether there is a significant enough quantity to require further action to protect the waterway and ecosystem. The following are broad categories and examples of how to determine the health of a stream.

Sediment

Sediment transport occurs when water in a stream carries sediment suspended in the water column or along the bottom of the stream. A stream naturally carries sediment, with high-volume waterways carrying greater loads. Too great of a sediment load is considered a pollutant and can lead to significant damage. Sources of soil **erosion** and soil loss include construction site runoff, agricultural runoff, streets and yards, and even sediment loss in the stream itself from damaging stormwater flows. High-volume discharges from unmanaged **impervious** surfaces can leave a stream unbalanced. A lack of vegetation to streambanks

WATERSHEDS & WATER MONITORING



Too much sediment can cause streams to widen and become warmer, reducing the amount of dissolved oxygen in the stream.

results in bank failure and increased sediment deposits.

As sediment settles out of the waterway, it covers stream and lake bottom habitat, harming the community that thrives in the riffles, runs, and pools of a natural channel and changing habitat availability. When sediment fills in the spaces between rocks and substrate, this is called "**embeddedness**." As streams receive more sediment, they also become shallower and tend to widen, slow down, and warm. Warm water holds less oxygen than cool water, leading to declines in dissolved oxygen. Sediment also carries adsorbed nutrients (attached to the sediment surface) and metals.

Nutrients

Nutrients, such as nitrogen and phosphorus and their various forms, are natural and needed for plant growth, but can be harmful in large quantities. Nutrients come from many sources including leachate from septic systems and fertilizer runoff from agriculture, golf courses, and commercial and residential properties. Nutrients are only a pollutant when excessive amounts result in a condition known as **nutrient enrichment**. Nutrient enrichment is evidenced by rapid growth of algae, or an **algal bloom**. When algae dies, bacteria that decompose these plants utilize the in-stream oxygen, causing **hypoxia** (low oxygen condition). The abnormal depletion of oxygen is a stressor for fish and other aquatic animals and can result in a fish kill.

Relating back to sediment, this chain reaction can more easily occur when streams have become shallower, more sluggish, and more easily warmed by the sun's rays.



An algal bloom is a sign of nutrient enrichment. When the algae dies, in-stream oxygen is used up by bacteria in the decomposition process, which can result in harmful hypoxic conditions.

Temperature

Temperature is an important measure of the health of a stream and its ability to serve as a functioning ecosystem. Water temperature has a significant effect on its inhabitants, where certain species such as trout require cold waters to reproduce. Warmer temperatures may increase fish vulnerability to disease and infection; water temperature also plays into the toxicity of ammonia in a stream.

Groundwater seepage and natural springs provide cold water to streams. Runoff that infiltrates through best management practices or riparian buffers similarly feeds cooler water to receiving waterways. However, where tree canopy and riparian buffers have been reduced, or stormwater flows from hot parking lots and roadways, this runoff becomes a thermal pollutant.

Bacteria

WATERSHEDS & WATER

MONITORING

Bacteria is an important measure of waterway health because of the inherent risk of swimming in waters that may be contaminated by viruses and harmful fecal-related waste. Sources of fecal-related bacteria to waterways include failing septic systems, wastewater failures and combined sewer overflows, illegal dumping, livestock, domestic pets, wildlife, waterfowl, and rodents. The result of an *Escherichia coliform* (*E. coli*) test only alerts the user to the presence of fecal-related bacteria in a waterway, but not the specific source. *E. coli* is an indicator species; the presence of *E. coli* indicates that the conditions in the stream are conducive to other possible pathogens as well.

Factors affecting bacteria levels include seasonal weather, stream flow, water temperature, distance from pollution sources, livestock management practices, wildlife activity, age of fecal material, sewage overflows, and rainfall. Bacteria can survive and grow in stream sediments for extended periods. Bacteria naturally die off with predation and UV degradation. Bacteria can settle to the stream bottom and be predated upon, or with nutrients and protection, populate. Because of these natural processes, bacteria sources are more difficult to trace, since distance from the source alone could alter the result.

Fortunately, methodologies including DNA source tracking are becoming more readily available. Quantitative polymerase chain reaction, or qPCR, is now the most trusted method of microbial source tracking, where DNA from specific species can be targeted in a water quality sample, amplified if present, and counted to assign a population density to quantifiable species. This can be a next step for resource managers in solving bacteria pollution in waterways.

2.3 GETTING TO KNOW YOUR WATERSHED

Watershed Driving Tour

With your watershed map in hand, you are ready to "hit the road." Taking the time to familiarize yourself with your watershed will be time well spent as you begin your monitoring program. Doing this at the onset of your project will give you a firm sense of what factors in your watershed may impact water quality. You should have at least two people in the car— one to drive safely and one to mark your map and take notes. It is helpful to go during a time of day when there is not a lot of traffic. Take your tour in daylight, so you don't miss structures that may not be visible at night.

What should you be observing and recording? The answer is ANYTHING that may affect your waterbody. Some suggested questions and notes to take are included on the next page, but this list is not exhaustive.

The information collected during your driving tour is for your use only. Collecting this information is especially important at the beginning of your monitoring effort, but as construction or other major events occur in your watershed, revisiting this "windshield survey" may be helpful to better understand your monitoring results and stream observations.

Walking the Stream

You have made a map, viewed your watershed and its characteristics in the <u>SC Watershed</u> <u>Atlas</u>, and conducted your windshield survey. Now is the time to choose the legally accessible point to carry out your monthly monitoring activities. Before you begin your sampling, walk upstream and downstream, looking for many of the same things you were looking for in the driving tour. Note areas of litter and trash dumping and make a note to clean that up at

your next visit (if you brought a bag, go ahead and clean it up now). If you find discarded tires, car parts, appliances, or other large trash items, contact your local government and partner with the town or county (Adopt-a-Highway), Palmetto Pride, and/or local Keep America Beautiful groups to schedule a larger cleanup. These groups will often provide bags, gloves, and other supplies to assist with litter collection efforts. You can also note suspicious activity and illegal dumping in the SC AAS Database.

Look for pastures, animal access to the stream, signs of animal activity in the stream and on stream banks, and areas of erosion. Take photos, and plan to do so regularly to have a visual log of changes to your stream. Never trespass where you do not have permission to be while doing this survey. Don't sample the same day as you do this stream walk, as you will have stirred up sediment that will affect your measurements.

 <u>Stormwater Infrastructure</u> - Is your watershed rural or urbanized? Are there ditches or storm drains along roads, or does stormwater sheetflow from roadways? Is the conveyance and its management effective or leading to visible erosion? <u>Sewer Infrastructure</u> - Do you see manholes and trunk lines indicative of sewer service? If not, the area may be served by individual septic systems or a mix of both. <u>Reservoirs, ponds and dams</u> - Are there large bodies of water that may serve as settling basins for sediment and associated pollutants? Are there dams? How are shorelines maintained? <u>Animals and bacteria</u> - What are your likely sources of bacteria in the watershed, if animal-derived? These may include livestock, high waterfowl populations at lakes and stormwater ponds, rookeries, or high populations of cats and dogs. <u>Golf courses</u> - Are there streams or drainage ditches within the course, and what is their management? Any ponds? Note geese and other waterfowl. <u>Row orops</u> - Is there evidence of good conservation practices such as grassed waterways, terraces, or contour oropping? Are streams lined with conservation buffers, or is the cong growing to the water's edge? <u>Residential areas</u> - Can you pinpoint where storm severs enter streams? Is there evidence of failing stormwater infrastructure, such as roads or ditches caving towards the direction of stormwater flow? <u>Construction Areas</u> - Depending on site size and proximity to waterway, there may be controls in place to keep sediment on site. Do you see silt fences, and if so, are they upright and intact? Is there very much sediment on the roadway at entrance and exit locations? 	FIELD NOTEBOOK
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CH.3: STREAM MONITORING WITH SC AAS



3.1 INTRODUCTION AND OBJECTIVES

The SC AAS Program offers trainings and monitoring resources for wadable freshwater streams in four protocols – Stream Habitat Assessment, Physical/Chemical Monitoring, Bacteria Monitoring, and Macroinvertebrate Monitoring. The parameters and frequency of data collection is unique to each.

PROTOCOL TYPE	PARAMETERS	MINIMUM FREQUENCY PER Active site
STREAM HABITAT ASSESSMENT	STREAM CONDITIONSRIPARIAN CONDITIONSHABITAT QUALITY AND QUANTITY	ONCE PER YEAR
PHYSICAL/CHEMICAL	 DISSOLVED OXYGEN AIR AND WATER TEMPERATURE PH TRANSPARENCY 	MONTHLY
BACTERIA	• E.COLI	MONTHLY
MACROINVERTEBRATE*	ORGANISM DIVERSITYORGANISM QUANTITYHABITAT CONDITIONS	TWICE PER YEAR

*Macroinvertebrate monitoring is a separate workshop and certification type.

WITH SC AAS

Each of these protocols include the recording of basic observations, including current and recent weather and precipitation, water clarity, color, and odor, presence of trash, conditions in and around the stream, and photos. These observations, while they may appear minor, are critical to connecting conditions to monitoring results, data trends, and stream management.

A note on photos: the SC AAS Database allows for three photos per monitoring event.

Take photos looking upstream and downstream and of any circumstances that you may want to document, such as sources of bacteria adjacent to the stream, severely failing streambank, riparian health, or discolored discharged or surface water. Photos must be JPG/JPEG or PNG files and no larger than 16MG to upload to the Database.

3.2 SELECTING YOUR MONITORING SITE

SC AAS encourages you to identify the site or sites that best meet your objectives, after first considering safety and access. In following our SC AAS Code of Ethics, be sure to seek written permission to monitor on private land, complete with a schedule of monitoring dates and what you will do while on the property.

Consider the following factors in your site's selection:

- Identify a site with easy access for you to carry equipment, and with a stable, clear location for you to set down bottles and other sampling supplies.
- Try to choose a site in a location convenient to you, like near your home, school, or work.
- Bridges most often provide easy access to a stream, though their presence modifies the natural stream setting; therefore, you may want to walk up or downstream to find a reach that is more representative of the waterway and its conditions. We recommend that you start upstream, if access allows.
- SC AAS seeks to provide data to the state on streams with active, year-round flow. Double check with landowners or county/city employees that the stream you have selected runs all year long.
- Your stream site must be wadable, but deep enough to allow you to collect samples without disturbing the sediment on the streambed. Be sure you can submerge bottles upright in an area with well-mixed, flowing water without hitting the stream bottom. Testing a water sample that includes disturbed and suspended sediment is not representative of the flowing water and should be discarded.
- If sampling near a lake, select a site on a stream that is upstream of the lake. Do not sample directly below a dam or other manmade structure/outflow.

3.3 RECORDING SITE OBSERVATIONS

All monitoring protocols include the recording of essential environmental conditions that set the stage for the monitoring results you will record. These data also aid in the State Team's ability to administer the program. From time spent monitoring and miles traveled to weather observations, these sections of your forms are equally important to the data you are recording for the stream. The following pages provide further explanations and resources for consistently collecting observations for all monitoring events.

WEATHER

Weather strongly influences the physical characteristics of water, and both short- and long-term weather conditions can greatly affect our streams. For example, a strong 4-hour spring rain may yield most of the stream's annual sediment and pollution load. Floods, droughts, or other climatic extremes can change the stream's physical and chemical characteristics dramatically (e.g., creating new stream channels or drainage patterns). As a rule of thumb, **water sampling should never be done during or immediately following a rain event. Wait at least 24 hours after rain, or until high flow has subsided, before sampling.**

TH SC AAS

- Weather can impact streams in several ways:
- Recent rains may increase nonpoint source pollution by increasing surface water runoff and • pollutant transport, which can decrease transparency and increase E. coli.
- Recent rains may dilute point source pollution. •
- Cloudy weather may limit plant photosynthesis and result in lower dissolved oxygen levels. •
- Wind may raise dissolved oxygen levels by increasing turbulence.
- Temperature affects many parameters, like the stream's ability to retain dissolved oxygen.

Reporting Technique

Report the weather conditions at the time of your stream assessment. Use the thermometer to measure air temperature before you use it for water temperature. Take measurements in the

shade. Report total rainfall for the previous 24 hours. Use your own rain gauge, check local weather sites, or refer to the CoCoRaHs website, www.cocorahs.org, for the nearest volunteer weather data.

WATER COLOR

Water color can provide immediate clues to a stream's condition.









Tannic



Oily Sheen





Milky/white



Gray

- No Color—Clear water does not necessarily mean clean water, but it could indicate low levels of dissolved or suspended substances.
- Brown/Muddy—Brown water is usually due to heavy sediment loads. Some South Carolina rivers more regularly run brown due to their soil types.
- Green—Green water is usually the result of excessive algae growth.
- Tannic—Blackish water is usually caused by natural processes of leaf decomposition. Pigments leached from decaying leaves can cause the water to appear murky.
- Milky/White—a milky appearance may be caused by salts in the water. It can also be a sign of illegal disposal by contractors or dischargers.
- Oily sheen—Oily sheens can be caused by petroleum or chemical pollution, or they may be natural by-products of decomposition. To tell the difference, poke the oily sheen with a stick. If the sheen swirls back together immediately, it is petroleum. If it breaks apart and does not flow back together, it is from bacteria, plant, or animal decomposition.
- Reddish—Reddish or orange colors are usually due to iron oxides.
- Gray—Gray water may be a result of natural or human-induced activities. Vegetation can produce surface-acting agents, or surfactants, which can cause surface foam. Human-induced surface foam may be an unnatural color (red, pink, blue, yellow, or orange) and have a fragrant smell. This foam is likely generated by household detergents and may be a sign of a failing septic drain field or illegal discharge.

Reporting Technique

Use a clear container or Whirl-pak bag to observe color in a water sample, as sometimes the stream may only appear a certain color due to the color of the streambed. Report observations based on the colors above.

In the SC AAS Database, any entry for "Other" will trigger an email alert to the SC AAS State Team, SCDES, and the local municipality or county, if they have agreed to receive such alerts.

WATER ODOR

Water odor, like water color, can provide immediate clues about potential problems in a stream:

- Gasoline—any petroleum or chemical smells may indicate serious pollution problems from a direct source, such as a factory, parking lot, or storm sewer runoff.
- Sewage/Manure—these smells can be common in the air, but should not be what our water smells like. It is important to determine whether the odor is coming from the water or air.
- Rotten egg—this odor may be caused by hydrogen sulfide gas, a by-product of **anaerobic decomposition**. This is a natural process that occurs in areas that have large quantities of organic matter and low levels of dissolved oxygen. It may be caused by excessive organic pollution.
- Fishy- this smell may be a sign of dead and decomposing fish in the stream.
- Chlorine- this smell may be a sign of pollution and will smell like a swimming pool.

Reporting Technique

Determine whether the water produces any of the odors described above. It's helpful to record this observation as soon as you arrive at your site, as odors may become less noticeable in a short time. To be sure an odor is coming from the stream and not the air, try smelling a water sample collected in a bottle.

In the SC AAS Database, any entry for "Other" will trigger an email alert to the SC AAS State Team, SCDES, and the local municipality or county, if they have agreed to receive such alerts.

CH.4: STREAM HABITAT ASSESSMENT PROTOCOL



4.1 INTRODUCTION

We have already established that the Stream Habitat Assessment allows one to track changes in the stream's stability, habitat quality, riparian conditions, and more. These qualitative characteristics can be documented in a quantitative way using this protocol. When conducting your Stream Habitat Assessment, use the appropriate form included in the Appendix of this Handbook.

Although Stream Habitat Assessments are designed to be completed once per year, you can use this assessment more frequently as site changes occur. As a steward of your waterway, you may want to conduct this assessment after significant events in your watershed such as before and after completion of a large industrial or commercial site, a flood or other catastrophic natural event, or new stormwater infrastructure placement. However often you choose, this protocol should always be completed with the assumption of

"leaf out" conditions so that riparian vegetation and stream shading can be compared independent of season.

Stream Reach

To properly use the Stream Habitat Assessment, you must determine your stream reach, which will be the same every time you conduct the assessment for each monitoring site.

QUICK NOTE

Leaf-out conditions assume that all healthy trees and shrubs are in their late spring to early fall show with all of their leaves. Your stream site is the location at which all freshwater data collection will occur. It also is the point where all bank full and active channel width measurements will be taken. You may want to mark this with flagging tape around a tree to remember the exact location.

The **reach** is the extent of stream for which you will assess and score. The reach should be about 12 times the channel width measured at your site. If your stream width is 15 feet, your reach would be 225 feet (15 ft x 12 = 225 ft). Your site is generally the approximate center of the reach, but you may instead need to determine your reach extent in either direction based on visibility, access, and conditions that are similar throughout. For example, the upstream reach may look generally the same on the left and right bank for a stretch, let's say 100 feet, until a landowner has obviously modified conditions in and around the stream. This change would demarcate the end of that reach, and you would instead look downstream from your site for the remaining 125 feet of your reach, if possible.

Stream Width

Regularly documenting the stream width at the same location provides information on how the stream may be changing and identify opportunities for restoration or mitigation of upstream impacts. You can record these measurements during monthly monitoring or during your annual Stream Habitat Assessment.

In SC AAS, there are two widths that we will record – "Bank Full" width and "Active Channel" width.

- **Bank Full Width** refers to the width of stream when it is at its most full condition, before it spills over to the floodplain.
- Active Channel Width is the width of stream where water is running on the day of monitoring.

A straight run works best for measuring width, but always remember to select a location that is stable, lacks obstructions (bridges, culverts, or crossings) and includes

pools or riffles within the reach. Use this as your location for measuring width each time.

A measuring tape or open reel measuring tape works best. This task is easiest done working with a partner, moving the tape from active to bankfull on each side to record the two measurements. If working alone, the reel should have a hook that can bite into or loop around a tree branch or root at your measurement location, as you cross to the other side of the river to find the distances.

Depth to Water Surface

Documenting depth to water from top of bank is another measure used to track a stream's changing morphology. Is the channel becoming more incised, with the top of bank getting further away from the surface of the water, reducing chances of the stream's access to its floodplain? Conversely, with measurements of stream width, do we see the water surface elevation staying relatively the same or rising, but channel getting wider? Streams naturally meander, especially in areas like the Sandhill ecoregion. Our ability to track these changes may allow scientists and hydrologists to intervene to address channel stabilization before a situation becomes exorbitant to repair or address.



4 STREAM HABITAT ASSESSMENT PROTOCOL

STREAM HABITAT ASSESSMENT PROTOCOL General Points to remember:

- Depth should only be recorded where and when there is safe access to the stream.
- Taking this measurement is most easily accomplished with a partner.
- The measurement should be taken from the lip of the stream bank at "Bank Full" height.
- In a stream that is naturally sloped, this measure may be more of an approximation, as one is measuring depth to water, not width of stream bank to water.

Stream Bottom Classification

The volunteer must first establish the type of stream being monitored in its natural condition – is it naturally a rocky bottom stream or a muddy/sandy bottom stream? This is a critical first step in completing the assessment protocols, as stream types are not compared equally for all

characteristics and will not have the same habitat opportunities for aquatic species.

4.2 HABITAT ASSESSMENT PARAMETERS

1. Epifaunal Substrate

Epifaunal substrate is the structures and materials on the stream bottom that provide surfaces for aquatic macroinvertebrates and fish to live. When observing your streambed, do you see roots and woody debris, leaf packs, cobbles, and/or undercut banks? If so, how abundant are these habitat types? Stable habitat cover is critical to support healthy populations of aquatic species, and thus, a variety and abundance of these types is rated highly in the Stream Habitat Assessment.

SO, WHAT FACTORS DETERMINE A STREAM'S STABILITY?

Channelization – Stream straightening, often seen in bulldozer widths, means faster moving water that may be incising (eroding the stream banks).

Soil types – The type of soil the stream channel is cutting through affects the strength of the bank. Soft loamy soils tend to produce eroding banks, whereas soil with more clay may be more stable. In the Stream Habitat Assessment, Channel Sinuosity is therefore only assessed for muddy/sandy bottom streams, where a greater meander is natural and not necessarily a sign of degrading conditions

Vegetation – A community of native wetland plants at the water's edge will prevent or slow bank erosion. Similarly, a mix of woody vegetation and grasses on the bank and in the floodplain leads to greater stability and opportunities for infiltration.

Foot paths - Trails and foot paths can create sluices for rainwater to flow downhill to the stream, eroding the foot path, increasing slope, and generally leading to unstable stream banks.

Livestock – Cattle can be especially destructive of streambanks and stream beds. Creatures of habit, livestock will create a "cow path" between pastures and to food and water sources.

Wildlife – South Carolina streambanks and river beds are being rooted and eroded by rapidly growing wild hog populations. This can be seen as stirred-up mud along the river, rooted wetland and adjacent agricultural plants, signs of wallowing in mud, and tracks. Wild hogs are not only a nuisance – they outcompete native animal species, damage ecosystem health, and carry disease. Noting their presence in your Stream Habitat Assessment is important for both potential sources of bacteria, as well as tracking population and damage throughout a watershed.

Infrastructure – Pipe outlets can cause erosion if not properly positioned, installed, or maintained. Document pipes and other infrastructure and its condition, if applicable, in your sampling reach.

2. Embeddedness

Embeddedness is the measure of and degree that larger particles (boulders or gravel) in a stream are surrounded or covered by fine sediment. Embeddedness is the result of excessive amounts of sediment settling on the streambed and filling the cracks and crevices that are macroinvertebrate habitat, and spaces where fish hide from prey, lay their eggs, and feed on their choice food. In this way, embeddedness reduces biodiversity of a stream.





Top: In this streambed composed of gravel and cobble, spaces between rocks are not filled with fine sediment. This stream is not embedded. Bottom: This stream is somewhat embedded, having more

spaces between rocks that are filled with fine sediment.

Using the Stream Habitat Assessment Form, you can score embeddedness based on the ability to see levels and sizes of various rocks and gravel in the streambed, or percentage of sediment that fills these spaces.

3. Riffle/Run/Pool

A variety of habitats within a stream usually enhances the diversity of aquatic life that you find there. Healthy streams show alternating pools, riffles, and runs:

- A **pool** has a relatively slow current and is usually found at a stream channel bend, upstream of riffles, and on the downstream side of obstructions such as boulders or fallen trees. The stream bottom in a pool is often bowlshaped and serves as excellent habitat for fish. It will be at least 1.5 times the average depth of your stream reach.
- A **riffle** is an area of the stream that has a swift current and water that is normally "bubbling" due to a rocky streambed. The water in this habitat type has relatively high dissolved oxygen levels from tumbling over and around the rocks. Riffles also typically have a high number of invertebrates.
- A **run** has a moderate current, medium depth, and smooth water surface. Runs can have diverse mixtures of aquatic life depending on the quality and quantity of the in-stream habitat (boulders, logs, root wads, leaf packs, etc.).

Sometimes stream channels have been modified artificially to improve drainage from surrounding land or to create passage across the channel, which can have lasting impacts on stream habitat health. Streams with long, continuous runs were often modified that way many decades or centuries ago, and are now restricted from meandering due to impervious surfaces, streambank controls, and infrastructure. In larger streams or rivers, pools are commonly the result of manmade structures or impoundments. Streams with little variety of habitat types will receive a lower score for this parameter.

4. Sediment Deposition

Sediment deposition evaluates how sediment is changing a stream's morphology. Islands and sandbars evolve as a result of heavy sediment loads in the stream, either due to erosion of the banks upstream, scouring of the streambed, poor filtration of stormwater runoff, or all of the above. Rapid and abundant sediment deposition can damage or destroy aquatic habitat and be a sign of overall unstable stream conditions. Comparing the presence and size of sandbars and islands between Stream Habitat Assessments is an important part of scoring this parameter.

STREAM HABITAT ASSESSMENT PROTOCOL

5. Aquatic Vegetation

Aquatic vegetation in and along the margins of streams can provide an important food source and rich habitat for aquatic macroinvertebrates; however, an overabundance of aquatic vegetation, including algae and submerged plants, can reduce habitat health. As discussed in Chapter 2, when algae dies, in-stream oxygen is used by bacteria that decompose plant materials, which can cause hypoxia. The rapid depletion of oxygen is a stressor for fish and other aquatic animals and can lead to fish mortality. Agricultural runoff, sewage spills, and other human activities can cause accelerated nutrient enrichment of waters, or **cultural eutrophication**, leading to the overgrowth of aquatic plants and algae.

When assessing this parameter, observe visible vegetation and algae, as well as any green, gray, or brown tint to the water that can be characteristic of an overabundance of vegetation. If algae is not visible, slippery rocks are a good indicator for its presence.

6. Channel Alteration

Channel alteration concerns human impacts to a stream's morphology and character. In both urban and rural areas, streams are sometimes modified or even rerouted to suit development and agricultural interests. Signs of historical channel alteration may be more obvious, such as banks lined with rip-rap or concrete, or the stream may have begun to recover and naturalize. Look for straight stream channels with no meanders, channels the width of a bulldozer, and any sign of unnatural materials used to stabilize banks.

7. Channel Sinuosity

Channel sinuosity is only assessed for muddy/sandy bottom streams, which have a greater tendency to

meander. All streams naturally meander, but this is often restricted by urbanization. When assessing this element, a greater proportion of bends and curves when compared to straight sections will yield a higher score.

8. Streambank Stability

The remaining parameters are evaluated by scoring the **left and right banks** separately. Determine left and right bank facing downstream, or in the direction of stream flow, as shown in the picture above. By evaluating and tracking a stream's stability, one can better understand and project if a stream will support a healthy



Determine left and right bank facing downstream.

aquatic community. For this assessment, it is important to note **streambank stability**, including vegetation whose roots and presence play a critical role in keeping streambanks stable long-term.

Streams require regular access to their natural **floodplain**, where high waters can spread out and be slowed by plants and topography. Think of the floodplain as the brakes on a bike; the bike picking up speed on the downhill will continue to accelerate without a change in topography or anything to slow it down. Without access to its floodplain, a stream similarly continues to pick up volume and velocity during and after storm events. This often leads to "hungry waters" that eat away at their own streambank, adding sediment to the stream or destabilizing roadways, bridges, and other infrastructure. This results in stream "scouring" or "incising." Stream incision occurs when a lack of access to the natural floodplain results in a drop in streambed elevation; this is commonly referred to as a "**degrading stream**." A healthy stream will naturally meander over time and within its floodplain, but a stable, mature stream will not move very rapidly. The presence of soil-holding vegetation protects the streambank from losing sediment, and with that, stability. Vegetation also plays other critical roles of shading the stream, adding habitat above the stream and in the stream (woody debris, leaf packs, logs), and slowing and infiltrating runoff.

9. Vegetative Protection

A diverse variety of vegetation and species helps to stabilize a stream and enrich the aquatic food web. **Vegetative protection** includes **canopy cover**, or the amount of shading of the stream by tree branches leaves, grasses, and shrubs that protects the stream and its

inhabitants from extreme fluctuations in temperature. Remember that

scoring should be completed assuming full "leaf out" conditions, allowing volunteers to conduct the assessment anytime of the year and still be consistent in vegetative scoring.

To complete this Stream Habitat Assessment, it is helpful to be able to identify common **invasive plant species**. Invasive plants are those that have been introduced to an area and created a breeding population which grows so successfully that it damages and restricts growth of native plants and agricultural crops. South Carolina's most common invasive species in riparian corridors are kudzu, Chinese privet, Chinese tallow tree, multiflora rose,

Japanese stiltgrass, Tree of Heaven, and wisteria. When species like these take root, you will often find a monoculture of plants within the riparian corridor.

There are many comprehensive resources available to learn about invasive plant species in the Southeast. For a quick reference guide, visit <u>www.invasive.org</u>, managed by the Center for Invasive Species and Ecosystem Health.

Documenting a variety of vegetation (versus a monoculture) is an important factor in ecosystem health and a healthy stream as some species hold soil together more than others, are more resilient to pests or disease, and add different benefits to the aquatic food web. A riparian zone with just one species, even if a native plant species, can leave a stream susceptible to future erosion, channel instability, or loss of habitat.

10. Riparian Vegetative Zone Width

Vegetated riparian zones can be highly effective at protecting stream health by filtering runoff and stabilizing banks and channels, while also providing habitat for wild animals. **Riparian vegetative zone** width simply looks at the width of the riparian buffer in relation to the width of the stream channel. Ideally, the riparian buffer on either side of the stream will be at least three times the width of the channel and filled with a variety of native species.

Trails and small-scale human activities within the buffer might not hinder the buffer's ability to do its job, but a trail right along the edge of the stream can be a significant disturbance as the trail puts pressure on the bank. Mowing or farming right to the edge of the stream equates to a significant disturbance as well.

Use the Stream Habitat Assessment Form in Appendix 1 to begin your data collection!

Vegetative protection plays an important role in regulating stream temperature.



4 STREAM HABITAT ASSESSMENT PROTOCOL

CHAPTER END

CH.5: PHYSICAL & CHEMICAL MONITORING PROTOCOL



5.1 INTRODUCTION

Physical and chemical monitoring of freshwater streams allows information to be gathered about specific water quality characteristics. In addition to basic visual observations and weather information, SC AAS recommends monitoring these core parameters monthly:

- Air temperature
- Water temperature
- Transparency
- pН
- **Dissolved oxygen (DO)**

If you choose to conduct freshwater stream monitoring, plan on sampling regularly – at least once per month, at the same time of day, and at the same location in a wadable stream with year-

QUICK NOTE

The SC AAS Freshwater Monitoring protocols included conductivity from 2015-2021. Those who were trained in this protocol may continue to monitor. Contact your Trainer if you need to return a conductivity meter.

round flow. Regular monitoring enables your chemical data to be compared over time, and water quality and environmental conditions can change throughout the day, so monitoring at approximately the same time of day is important. Also, chemical testing during or immediately after a rain may produce very different results than during dry conditions, so it's best to wait at least 24 hours after rain to sample.

As you explore your stream's water chemistry, it is important to understand that water chemistry is very complex and that natural variation in some parameters is not unusual, but actually the norm. The following are some examples of how environmental conditions can influence water chemistry:

- Time of Day DO levels rise during sunlight hours due to increased photosynthesis in aquatic plants and algae, and decrease overnight when photosynthesis is not occurring.
- Weather Runoff from heavy rains can transport pollutants, especially sediment, to streams.
- Physical Influences Decreased canopy cover results in solar warming of the water, which can decrease DO levels.

5.2 CONTENTS OF THE CHEMICAL MONITORING KIT

The list in Appendix 2 includes everything you should need to conduct your SC AAS Freshwater Monitoring. In addition to the necessary sampling/testing supplies, we recommend bringing this Handbook, a trash bag to pick up litter, a First Aid Kit, and waders, boots, or old tennis shoes.

Reagents used in measuring pH and DO have expiration dates. Please be sure that your reagents are not expired before monitoring. Results found using expired reagents are not trustworthy for reporting and sharing, and therefore, are blocked from being entered into the database. If you need to replenish your reagents in your SC AAS monitoring kit, please contact your Trainer for guidance or refer to the kit supply list in Appendix 2.



Photo c/o Anaston Porter



Hang the thermometer in the shade to measure air temperature first.

5.3 RECORDING AIR AND WATER TEMPERATURE

Air temperature is taken first, as a reference point for water temperature. Water temperature is one factor in determining which species may or may not be present in the system. Temperature affects feeding, reproduction, and the metabolism of aquatic animals. A week or two of high temperatures may make a stream unsuitable for sensitive aquatic organisms, even though temperatures are within tolerable levels throughout the rest of the year. Not only do different species have different requirements, optimum habitat temperatures may change for each stage of life. Fish larvae and eggs usually have narrower temperature requirements than adult fish. PHYSICAL & CHEMICAL

Temperature Monitoring Technique

- Use a thermometer protected by a plastic or metal case
- Measure air and water temperature in the shade.
- Take air temperature before water temperature. (Tip: hang the thermometer in a tree when you arrive at your site!)
- Let the thermometer stabilize for 60 seconds in the air/under the water before reading.
- Fully submerge the thermometer in the stream for water temperature.
- Record temperatures in degrees Celsius.
- Hold the thermometer at its top or by a string.
- Check your thermometer for bubbles before sampling. If the liquid appears separated, temperature readings will not be accurate. You can purchase a new thermometer (see Appendix 2) or notify the owner of your kit, if borrowing from a Hub.

State Standards

The water temperature of all free flowing freshwaters shall not be increased more than 5°F (2.8°C) above natural temperature conditions (unaffected by anthropogenic sources of pollution) and shall not exceed a maximum of 90°F (32.2°C) as a result of the discharge of heated liquids unless different site-specific criteria have been established. In trout waters, temperature shall not vary from levels existing under natural conditions, unless determined that some other temperature shall protect classified uses.

5.4 TRANSPARENCY

Transparency is the clearness of water, also known as clarity. Color, algae, phytoplankton, and suspended sediments can all impact transparency results. **Turbidity** is the measure of the cloudiness of water, measuring how suspended material affects light penetration into the water column. Turbidity can impact photosynthesis and water temperature, and can indicate unmanaged runoff to the waterway. Transparency and turbidity are inversely related, so as transparency decreases, turbidity increases. In other words, the higher the turbidity,

the less transparent the water is. SC AAS uses the Transparency Tube to record transparency, from which we can then approximate turbidity.

Many "pollutants" in a stream occur naturally and are only considered a pollutant when their presence is impairing a waterway's ability to maintain life, or when they are a potential threat to users. This is the case with sediment. In fact, sediment is one of the top nonpoint sources of freshwater pollution!

Streams naturally carry sediment, but as they receive more sediment than their flow can transport, they become shallower and tend to widen, slow down, and get warmer. Warm water holds less oxygen than cool water, leading to declines in DO. Sediment also carries adsorbed nutrients (nutrients attached to their surface) and metals. Finally, sediment can disrupt stream habitat and clog the gills of fish.

0	32.0
1	33.8
2	35.6
3	37.4
4	39.2
5	41.0
6	42.8
7	44.6
8	46.4
9	48.2
10	50.0
11	51.8
12	53.6
13	55.4
14	57.2
15	59.0
16	60.8
17	62.6
18	64.4
19	66.2
20	68.0
21	69.8
22	71.6
23	73.4
24	75.2
25	77.0
26	78.8
27	80.6
28	82.4
29	84.2
30	86.0
31	87.8
32	89.6
33	91.4
34	93.2
35	95.0

Sources of sediment include construction site runoff, farm field runoff, urban runoff from streets, ditches, and yards, and even sediment loss in the stream itself. High volume discharges from unmanaged impervious surfaces can leave a stream unbalanced. A lack of vegetation on streambanks results in bank failure and increased sediment deposits.

The first sign of decreased transparency/increased turbidity at your site is often a brownish water color, such as after rain events. However, these conditions may not always correlate with precipitation. Regular monitoring during normal conditions is important to establish baseline levels and help identify any fluctuation. Assigning a numeric measure related to sediment load allows us to record imbalances in our rivers, and enables resource managers to determine if further action needs to be taken to protect a waterway.

State Standards

There is no SC state standard for transparency. To meet state standards for turbidity, most freshwater streams must not exceed 50 Nepehlometric Turbidity units (NTUs), provided existing uses are maintained. In special trout waters (Upstate and Lower Saluda), turbidity must not exceed 10 NTUs or 10% above natural conditions, provided uses are maintained.

Transparency Monitoring Protocol

To measure transparency, we use a transparency tube. The transparency tube is filled with stream water, which is then slowly released from the bottom until you are able to decipher the black from white quadrants of the Secchi disk inside the tube.

- 1. Approach your sample site from downstream. Be sure that you are not stirring up bottom sediments before you fill the tube.
- 2. If flow and depth allow, directly submerge your tube until it is full. Otherwise, use a bucket to collect your sample from an area of undisturbed flow and fill the tube with this water sample.
- 3. Cover the top with your hand and invert the tube a couple of times to be sure the sample is well-mixed.
- 4. Hold the tube vertically on a solid surface, such as a rock or sidewalk. Perform this inspection in the shade, without a hat or sunglasses.
- 5. Look straight down into the tube and press down on the tube to slowly release sample water. Slow down as you see the black and white secchi disk come into view.
- 6. When you can you can just begin to differentiate the black and white quadrants, stop releasing water. Look at the side ruler to take your transparency reading in centimeters.

When this transparency value is entered into the SC AAS database, it will be converted to turbidity for you with the unit NTUs. A turbidity conversion chart can be found at <u>www.scadoptastream.org</u>.





Look straight down into the tube while pushing down to release water from the base.

5.5 PH MONITORING

pH is a measure of how acidic or basic water is and is measured in pH units on a scale of 0 to 14. A pH of 7 is neutral (distilled water), while a pH greater than 7 is basic/alkaline, and a pH less than 7 is acidic. The pH scale is logarithmic, so every one unit of change in pH actually represents a tenfold change in acidity. A pH of 6 is ten times more acidic than a pH of 7; a pH of 5 is 100 times more acidic than a pH of 7.

The pH of stream water is influenced by the concentration of acids in rain and the types of soils and bedrock in the watershed. The typical rainfall in the U.S. is slightly acidic, with a pH ranging from 5.0 to 5.6. As rainwater falls, carbon dioxide from the atmosphere dissolves into weak carbonic acid and thereby lowering the pH of the precipitation.

it, forming a weak carbonic acid and thereby lowering the pH of the precipitation.

Low pH levels (acidic water) can have a harmful impact on the health of aquatic communities. Very acidic water or acid rain can allow toxic substances such as ammonia and heavy metals to leach from our soils and possibly be taken up by aquatic plants and animals in a process called bioaccumulation.

Most aquatic organisms require habitats with a pH range of 6.5 to 8.5. Extremely high or low pH values are quite rare in South Carolina. Most values that exceed 9.0 (basic) are caused by excessive algal growth, a sign of nutrient enrichment. Very low (acidic) pH readings are generally near point sources of pollution, however, there are surface waters in South Carolina's coastal plain where pH may be at the low end of or below the state standard range, which is the natural condition of these waterways due to tannins from leaves.

The pH of water is measured by adding a reagent to a sample of water, which reacts with the pH of the sample to produce a color. The color of the sample is then matched to a color comparator to determine pH.

State Standards

PHYSICAL & CHEMICAL MONITORING PROTOCO

In the vast majority of freshwater streams in South Carolina, pH levels should fall between 6.0 and 8.5

to meet South Carolina state standards. In special trout waters (Upstate and Lower Saluda) pH levels should fall between 6.0 and 8.0. Specially designated areas of the Coastal Plain (blackwater) will have lower pH levels due to natural conditions.

pH Monitoring Protocol

Instructions for the LaMotte Code 5858-01 (new model):

- 1. Check expiration dates of chemical reagents.
- 2. Put on gloves and eye protection before beginning the test.
- 3. Rinse the two plastic tubes two times with stream water.
- 4. Fill each tube to the 10 mL line with stream water.
- 5. Add 10 drops of the pH wide range indicator (holding indicator bottle vertical). Cap and gently invert the sample several times to ensure mixing.
- 6. Insert the wide range pH Octa-Slide Bar 2 into the Octa-Slide 2 viewer.
- 7. Insert pH test tube into Octa-Slide 2 viewer.
- 8. Match sample color to a color standard and record pH 0.25 standard units.
- 9. Discard processed samples into the waste container.



It's often helpful to use a white background when comparing your pH sample to the Octa-Slide bar.

Duplicate precision rule for pH: The two samples must be within \pm 0.25 standard units. If your two sample results differ by more than 0.25, resample until another sample results in a pH value within this acceptable range. Both final results must be recorded in the database.

Remember to always discharge the analyzed sample into the waste bottle and not into the waterway or on the ground. Rinse tubes with water to clean. Remember to put the caps back on chemicals.

5.6 DO MONITORING

Like land organisms, aquatic animals need oxygen to live. Fish, invertebrates, plants, and aerobic bacteria all require oxygen for respiration.

Sources of DO

Oxygen dissolves readily into water from the atmosphere at the surface until the water is full, or saturated. Once dissolved in water, the oxygen diffuses very slowly, and distribution depends on the movement of aerated water by turbulence and currents caused by wind, water flow, and thermal upwelling. Aquatic plants, algae, and phytoplankton produce oxygen during photosynthesis.

DO Capacity of Water

The DO capacity of water is limited by the temperature of the water. As water temperature changes, the highest potential DO level changes.

- At 0°C the saturation point for DO is 14.6 mg/L.
- At 32°C the saturation point for DO is 7.6 mg/L.

The temperature effect is compounded by the fact that living organisms increase their activity in warm water, requiring more oxygen to support their metabolism. Critically low oxygen levels often occur during the warmer summer months when capacity decreases and oxygen demand increases. This is often caused by the respiration of algae or by decaying of organic material.

Significant Levels

The amount of oxygen required by an aquatic organism varies according to species and stage of life.

- DO levels below 3 mg/L are stressful to most aquatic organisms.
- DO levels below 2 or 1 mg/L will not support fish; levels of 5 to 6 mg/L are usually required for growth and activity.
- Fish and invertebrates that can move will leave areas with low DO and move to higher level areas.

Interpreting Your DO Results

A low DO level indicates a demand on oxygen in the system. Pollutants, including inadequately treated sewage or decaying natural organic material, can cause such a demand. Organic materials accumulate in bottom sediments and support microorganisms (including bacteria), which consume oxygen as they break down the materials. Some wastes and pollutants produce direct chemical demands on oxygen in the water. In ponds or impoundments, dense populations of active fish can deplete DO levels. In areas of dense algae, DO levels may drop at night or during cloudy weather due to the net consumption of DO by aquatic plant respiration. Note that there are surface waters in South Carolina's coastal plain where DO may be very low, which is the natural condition of these waterways.



High DO levels can be found where stream turbulence or choppy conditions (such as riffles) increase natural aeration by increasing the water surface area and trapping air under cascading water. On sunny days, high DO levels occur in areas of dense algae or submerged aquatic vegetation due to photosynthesis. In these areas, the lowest DO levels occur just before sunrise each morning, with the highest levels just after noon.

State Standards

For the majority of South Carolina's freshwater streams, DO levels must average 5 mg/L and be no less than 4 mg/L to meet state standards. In trout waters (Upstate and Lower Saluda), DO levels must be no less than 6 mg/L with no daily average. Some specially designated waters of the Coastal Plain have lower DO levels due to natural conditions.

DO Monitoring Protocol

DO is measured using the Winkler Titration Method. A sample bottle is rinsed twice with stream water and then filled completely. Cap the bottle under water so that no air is present in the sample. Reagents are added to produce a 'fixed' solution, meaning the DO content cannot be influenced by external sources or changes. This fixed solution is then titrated until it reaches the 'endpoint' where the color of the solution changes to clear. The level of the remaining liquid in the direct-read titrator corresponds to the DO level in the sample.

Take two samples for duplicate precision and follow the steps below for both tests.

Duplicate precision rule for DO: The two samples must be within ±0.6 mg/L.

If the results are not within duplicate precision of each other, run an additional tests until two are within that range. Do not stand over the stream while conducting this test. It is also best to conduct the test while standing to avoid dripping chemicals on yourself. Sample bottles Titrators 1 2 4 3 5 Titration vials Thermometer

Contents of the dissolved oxygen test kit.

Before you begin:

- Check expiration dates on chemicals prior to sampling.
- Wear safety goggles and gloves while completing this procedure.
- Keep a supply of paper towels on hand to clean any spills right away.
- Rinse your sample bottles twice with stream water.
- Collect samples facing upstream and cap bottles underwater to be sure they are completely filled with water and no air bubbles are present.
- Follow the steps in the order they are written. It's best to process one sample at a time or have a partner process the second sample to avoid accidentally mixing test components.
- Always discharge analyzed stream water to the waste container and not on the ground or in the waterway.

Step 1: Do not have the cap off the samples for any longer than necessary!

- a. Holding the reagent bottle completely upside down, add 8 drops of Manganous Sulfate solution (labeled "1" on bottle) to each sample.
- b. Holding the reagent bottle completely upside down, add 8 drops of *H* Alkaline Potassium Iodide Azide (labeled "2" on bottle) to each sample.



Hold the chemicals completely upsidedown to maintain consistent drop size.

d. Let the floc settle until the top half of the sample bottle is clear.

Step 2: Be careful when using this chemical and do not allow kids to help with this step! Add 8 drops of Sulfuric Acid 1:1 (red cap, labeled "3" on bottle). Cap and gently invert the bottle to mix. The solution will turn from cloudy to translucent and a burnt orange color. If you still see some dark solids in the solution, add 1 more drop to each bottle. Your samples are now "fixed," meaning the oxygen level in the bottles can't change if you take the caps off.

Step 3: Rinse the titration vial twice with your fixed solution (one vial for each sample), then pour the fixed solution to the 20 mL mark.

Step 4: Fill the titrator (plastic syringe with pink tip) by putting the tip of the titrator into the hole in the top of the titrating solution Sodium Thiosulfate 0.025N (labeled "4" on bottle). Turn the bottle upside down and slowly pull back on the syringe plunger to draw some of the solution into the titrator. Push in the plunger to expel any air bubbles. Draw back the plunger again until the tip on the bottom of the plunger is well past the zero (0) mark on the scale on the titrator. Once all air bubbles are expelled, push the titrator so that the solution is exactly at the 0 ppm mark. *Note: DO is recorded in mg/L or ppm. These units are interchangeable for this measurement.*

Step 5: Turn everything right side up. Make sure the large ring on the plunger of the titrator is still at the 0 mark and there are no air bubbles. Repeat these steps if air bubbles are present.

Remove the titrator from the sodium thiosulfate bottle.

Step 6: Put the tip of the titrator into the opening on the plastic cap of the titration vial that contains your fixed sample.

Step 7: Titrate the sample: Add the titrating solution one drop at a time by gently pushing the plunger. Swirl the solution between each drop until the sample has turned pale yellow. If your solution is already pale yellow, skip this step. If your solution is colorless, you have 0 mg/L DO or you may have added too many drops (proceed to step 8 for confirmation).

Step 8: Pop off the plastic cap from the titration vial with the titrator still in the cap without moving the plunger. Add 8 drops of starch indicator solution (labeled "5" on bottle) to the vial. The sample should now turn dark in color.

Step 9: Continue titration after capping the vial, adding one drop at a time and swirling the solution between each drop. Immediately stop adding drops of sodium thiosulfate when the solution turns colorless. If the color change is not complete by the time the plunger tip reaches the bottom of the scale on the titrator, refill the titrator with sodium thiosulfate and continue the titration. Add both titration amounts together for the final test results (one full syringe is 10 ppm).

Step 10: Read the test result directly from where the scale intersects the ring of the plunger. The titrator is marked at 0.2 ppm increments. For example, if the titrator ring is touching the third line below the line marked "4" the DO result would be 4.6 mg/L. (If the titrator has been refilled once before, the result would be 14.6 mg/L DO.)

Step 11: Check duplicate precision: If the results are more than 0.6 mg/L apart between the two tests, repeat the test again until two results are within 0.6 mg/L of each other. Only report the two final readings in the database.

Titrator at 0 ppm.

MONITORING PROTOCOL



Modified from LaMotte, Dissolved Oxygen Water Quality Test Kit Instruction Manual, Code 5860-01.

Part 2 - Adding the Reagents REMINDER: Check expiration dates on chemicals.



NOTE: At this point the sample has been "fixed" and contact between the sample and the atmosphere will not affect the test result. Samples may be held at this point and titrated later.

*WARNING: Reagents marked with an * are considered to be potential health hazards.

Modified from LaMotte, Dissolved Oxygen Water Quality Test Kit Instruction Manual, Code 5860-01.

PHYSICAL & CHEMICAL

PHYSICAL & CHEMICAL MONITORING PROTOCOL

Part 3 - The Titration



NOTE: If small air bubbles appear in the titrator barrel, expel them by partially filling the barrel and pumping the titration solution back into the reagent container. Repeat until bubble disappears.



**Don't forget DUPLICATE PRECISION. Refer to chapter 5 in the SC Adopt-a-Stream Handbook for more information and FAQs.

Modified from LaMotte, Dissolved Oxygen Water Quality Test Kit Instruction Manual, Code 5860-01.



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Read the test result directly from the scale where the large ring on the Titrator meets the Titrator barrel. Record as ppm Dissolved Oxygen. Each minor division on the Titrator scale equals 0.2 ppm.

NOTE: When testing is complete, discard the titrating solution in the Titrator. Rinse Titrator and titration tube thoroughly. DO NOT remove plunger or adapter tip.



Modified from LaMotte, Dissolved Oxygen Water Quality Test Kit Instruction Manual, Code 5860-01.

PHYSICAL & CHEMICAL

5.7 COMMON QUESTIONS ABOUT ANALYZING PH & DISSOLVED OXYGEN

This assumes that you have collected your water sample(s) and have capped the bottle(s).

Should I pour off any of the water in my DO sample bottle before I add the reagents? NO! If you pour off some water you are introducing air (and oxygen). When you cap the bottle and shake it, this oxygen can cause erroneously high results. Put the bottle on a paper

towel if necessary to catch any water that spills over when you add the reagents. The reagents are more dense than the water and will sink to the bottom. Any overspill liquid is just water from your sample.

What should I do if reagents are expired?

If any of your reagents are expired, do not use them to perform this test. If your kit is borrowed from a Hub or Trainer, let them know that the chemicals are expired when you return them. If you own and maintain your kit, refer to Appendix 2 for a list of kit supplies and where to purchase refills. Expired manganous sulfate, sodium thiosulfate, and starch indicator can be flushed down a drain with plenty of water. Do not do this if you are on a septic tank. Alkaline potassium iodide, sulfuric acid, and pH wide range indicator must be taken to a local recycling center when they are accepting hazardous waste.

How should I hold the dropper bottles to dispense each reagent?

Hold the dropper bottles completely upside-down. This ensures a uniform drop size. The liquid reagents will not come out until you squeeze the bottle.

Sometimes after I add the eight drops of sulfuric acid (DO) some brown particles remain. Is this OK?

The brown particles should be dissolved before you continue with your test. First, try shaking the sample bottle quite hard to see if they dissolve. If this doesn't work add one more drop of sulfuric acid (red capped bottle). Occasionally in water with an algae bloom there may be some organic matter present in your sample. This won't dissolve. You should be able to tell the difference between this and the chemical particles.

What does it mean by saying that the DO sample is "fixed"?

In a practical sense it means that contact with atmospheric oxygen will not affect your test results. Fixed samples may be stored up to eight hours, if kept refrigerated and in the dark. The chemical reactions that occur in this analysis are explained after these questions.

My DO water sample is pale yellow right after it is fixed. Do I still have to see it get lighter before I add the starch indicator?

If your water sample is already a pale yellow after it is fixed, add the starch indicator before you begin your titration. If your sample is completely colorless after it is fixed and remains that way after you add the indicator this means that there is no DO in your sample. If this is the case, you might want to check the DO content of water from another source or your tap just to make sure that the reagents in your kit are still functioning properly.

How many times should I run the chemical tests on my water samples?

You should run the pH and DO tests at least twice on two samples for each. If the results are outside the duplicate precision range (+/- 0.25 for pH and +/- 0.6 ppm for DO), perform the test again until two samples are within the correct range of each other. For DO, you can first try retesting from your leftover fixed samples, as an error may have occurred in the titration process to cause the discrepancy.

What should I do with any leftover sodium thiosulfate in the DO syringe?

Discard any remaining sodium thiosulfate in your waste container. Do not put it back into the bottle it came from.

What do I do with chemical waste?

If you are on municipal water, you can flush the contents of your waste bottle down the drain with plenty of water. Do NOT do this if you are on a septic system! Simply return your waste to your kit loan location or contact your trainer for assistance.

What do I do with expired chemicals?

Expired Manganous Sulfate solution (DO #1), Sodium Thiosulfate (DO #4), and Starch Indicator solution (DO #5) can be flushed down the drain (NOT on septic) with copious volumes of water. If you are on a septic system, you can return these to your trainer or kit loan location.

The Alkaline Potassium Iodide Azide (DO #2), Sulfuric Acid (DO #3), and pH Wide Range Indicator are hazardous waste and must be disposed of at your local recycling center on a day when they are accepting hazardous waste. You can also return these to your trainer or kit loan location to be disposed of.

How do I clean my kit?

Simply rinse pH sample tubes, DO bottles, and titration tubes in the sink. Throw away any used gloves and paper towels, and dispose of chemical waste as described above.



BACTERIA MONITORING PROTOCOL

CH.6: BACTERIA MONITORING PROTOCOL



6.1 INTRODUCTION

Bacteria are microscopic, single-celled organisms. They are so small that five million could be placed on the head of a pin. Under favorable conditions, they can reproduce rapidly and can form colonies that are visible without magnification. Bacteria can utilize a large variety of habitats and can survive and adapt to almost all conditions present on planet earth. They have been so successful that they are the most numerous life forms on the planet. Most bacteria are beneficial and responsible for important environmental processes such as decomposition, nutrient cycling, and the breakdown of environmental toxins. Some bacteria, however, are **pathogenic** (disease causing) and may result in human health issues.

Coliform bacteria are members of the Enterobacteriaceae family. While some coliform bacteria can be naturally found in soil, the types of coliform bacteria that live in the intestinal tract of warm-blooded animals and are present in animal and human waste are called **fecal coliform** bacteria. *Escherichia coli (E. coli)* is one subgroup of fecal coliform bacteria. Even within this species, there are numerous different strains, some of which can be pathogenic.

What are Indicator Bacteria?

Trying to detect disease-causing bacteria and other pathogens in water requires considerable training, time, and expense. The US EPA recommends *E. coli* bacteria as a good indicator organism of fecal contamination in freshwater because they are associated with warm-blooded animal wastes, are found in greater numbers, and are less risky to culture in the lab than other pathogens. However, their presence does not necessarily

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mean that other pathogens are present, but rather indicates a potential risk to human health. Monitoring for these indicator organisms is an easy and economical method for citizens or professionals to assess health risks due to bacterial contamination of surface waters. If bacterial contamination of surface water is found, other disease-causing organisms such as viruses and protozoans may also be present and pose a health threat.

What Risks Do Bacteria Pose to Human Health?

Volunteer Monitors should be aware of the risk that high bacteria levels may pose to human health. The higher the bacteria levels, the greater the potential health risk for 1) gastroenteritis, a condition indicated by vomiting, diarrhea, fever, nausea, and stomachache; and 2) respiratory, eye, ear, nose, throat, and skin infections. While *E. coli* by itself is not generally

a cause for alarm, excessive levels of *E. coli* may indicate the presence of a pathogenic strain of *E. coli*, *Salmonella*, and *Shigella* (which can cause gastrointestinal illnesses), *Psuedomonas aeruginosa* (which can cause swimmer's ear or dermatitis), protozoans such as *Cryptosporidium* and *Giardia* and viruses such as hepatitis A. SCDES standards and US EPA recommended *E. coli* criteria are based on the association of illness

QUICK NOTE

The 3M Petrifilm method used in SC AAS measures *E. coli* in CFU (colony forming units) per 100 ml, while the SCDES standard is 349 MPN (most probable number) per 100 ml. This is a result of different testing methods. CFU counts visible bacteria colonies on the plate, while MPN is a statistical measure of growth.

rates of swimmers with E. coli concentrations or "contact recreation standards."

How do Bacteria Get into Streams and Rivers?

E. coli in waterways can originate from the intestinal tracts of both humans and other warm-blooded animals, such as dogs, cats, livestock, and wildlife. In urban watersheds, fecal indicator bacteria are significantly correlated with human density (Frenzel and Couvillion, 2002). Possible animal sources of fecal coliform bacteria include cattle in streams, land application of animal waste, dairy operations, poultry operations, hobby horse farms, dog and cat waste from parks, lawns, streets and wildlife such as geese, pigeons, ducks, deer and raccoons.

Fecal material as well as other pollutants can be transported to waterways through runoff from rain events. How quickly pollutants are transported partially depends on the type of land use and management of runoff.

Human sources of fecal matter:

- failing septic tanks
- leaking sewer lines
- wastewater treatment plants
- sewer overflows
- land application of biosolids
- boat discharges

Grasses and vegetated land tend to soak up rainfall, thereby increasing infiltration into the ground and reducing runoff to waterways. Developed lands such as streets, rooftops, sidewalks, parking lots and driveways create more impervious surfaces thereby increasing runoff. Lands that support animals such as dogs, cats, cattle, hogs or horses can also be a source of bacteria, particularly if animals enter the water for drinking or if heavy rains wash manure from the land into receiving waters.

Another source of bacteria in stream waters originates from point sources, such as the discharge of pollutants through a pipe. Bacteria can enter waterways from broken pipes, illicit connections, and stormwater outfalls. Additionally, large rain events, power failures, or technical problems can cause wastewater treatment plants to discharge partially treated sewage directly into rivers and streams due to the excessive volume of water entering the plant.

BACTERIA MONITORING PROTOCOL

6.2 BACTERIA SAMPLING CONSIDERATIONS

When to Sample

SC AAS recommends that sampling for *E. coli* should be done at least monthly during dry weather conditions, and at the same time of day each month. These factors help in the comparison of your data over time. The number of times that you will need to sample varies and depends on your goals. The more you sample, the better information you will have when interpreting your data. Remember to collect samples below riffles or in flowing water. It's recommended that bacteria sample collection be your last step before leaving your site to avoid prolonged exposure to UV light or heat.

Following SC AAS protocols, volunteer monitors process their samples at home using 3M Petrifilm[™] plates and an incubator. Many studies have shown that the 3M Petrifilm[™] method is as effective as professional lab methods and is more practical and cost efficient for the volunteer monitor.

6.3 BACTERIA MONITORING KIT

The list in Appendix 2 includes everything you should need to conduct your SC AAS Freshwater Monitoring. In addition to the necessary bacteria sampling/testing supplies, we recommend bringing this Handbook, a trash bag to pick up litter, a First Aid Kit, and waders, boots, or old tennis shoes.

Petrifilm plates for *E. coli* testing have expiration dates. Please be sure your plates are not expired before monitoring. If you need to replenish any supplies in your monitoring kit, please contact your Trainer for guidance or refer to Appendix 2.

QUICK NOTE

Practice using the pipette to get used to the two "stops". When depressing the plunger, the first point at which you feel resistance is the first "stop". Holding the plunger at this point, place the tip of the pipette in water and slowly release. This collects exactly 1 mL of water in the pipette tip. To dispense the water, push the plunger all the way down to the second "stop". This ensures the full sample volume in the pipette tip is released.



Practice finding the two pipette "stops" to collect and release sample water from the sterile tip (blue).

6.4 BACTERIA MONITORING PROTOCOL

Prior to sample collection, here are some tips to consider, which are also noted in the directions that follow:

- Unopened packs of 3M Petrifilm[™] plates should be stored flat at or below 8°C (46°F). Use before expiration date on package. Allow pouches to reach room temperature before opening. Allow frozen plates to thaw to room temperature (only takes a few minutes) before plating samples. To seal an opened pouch, fold end over and tape shut. To prevent exposure to moisture, do not refrigerate opened pouches. Store resealed pouches in a sealed container in the freezer until the expiration date OR in a cool, dry place for up to one month. Avoid exposure of plates to temperatures >25°C (>77°F) and/or relative humidity >50%.
- The incubator typically used in SC AAS kits is made of styrofoam. To reach 35°C for incubation, you may choose to plug in the incubator in a safe place before sampling.

Directions for Bacteria Monitoring Using 3M Petrifilm[™] Step 1: Preparing the blank/control sample

For each sampling event (a day of sampling up to 10 sites), the volunteer shall fill a Whirlpak® bag with tap water from home to serve as the blank/control. Prior to filling the control sample bag, label the Whirl-pak® bag as "Blank", with date and time, and the sample collector.

Having a field blank when you sample is necessary to serve as a control. A control will ensure that you are practicing a sterile technique that prevents contamination. If you are sampling at more than one site, prepare one blank for every 10 sites. The blank is then plated and analyzed with the stream samples in the lab. Lab analysis of the blank should result in a zero reading for bacteria. If it is contaminated, you will need to discard all samples; no data can be submitted for these samples. Resample, and if your control sample again shows bacteria growth, contact your water supplier, or if you use private well water, disinfect the well. For information on how to disinfect your well, go to this link: <u>https://des.sc.gov/programs/bureau-water/residential-wells/privatedrinking-water-wells</u>

- 1. Before heading to the field, correctly label 1 Whirl-pak® bag with a permanent marker for the blank/control (1 blank per 10 sample sites).
- 2. Remove the perforated seal from the top of the Whirl- pak® bag. *IMPORTANT! Do not touch the inside of the Whirl-pak® as this could contaminate your sample and alter the results.*
- 3. Use the two small white tabs to pull open the bag.
- 4. Fill the Whirl-pak® bag 2/3 full with tap water.
- 5. Grab the ends of the twist ties and "whirl" or spin the bag tight. Cross the twist ties to close the bag.
- 6. Make sure the bag is closed securely by inverting the bag to test the seal (no water leaks out).
- 7. Immediately place the Whirl-pak® bag into a properly disinfected cooler with ice and take it with you throughout your sampling event.

Step 2: Collecting site sample in the field

Use one Whirl-pak[®] bag to collect a sample at each site you monitor.

- 1. Label an unopened Whirl-pak® bag using a permanent marker with the sample/site name, date, and time.
- 2. Put on latex gloves and remove the perforated seal from the top of the Whirl-pak® bag. *IMPORTANT! Do not touch the inside of the Whirl-pak® as this will contaminate your sample and could alter the results.*
- 3. Use the two small white tabs to open the bag.
- 4. While holding the twist ties, place the bag in the water at mid-stream, mid-depth or in a well-mixed area and allow the water to flow into the bag. Remember to collect the water sample (at least wrist deep) upstream of where you are standing. Fill the bag with water up to 2/3 full. Discard and refill if sediment gets in the bag.
- 5. Grab the ends of the twist ties and "whirl" or spin the bag until tight. Cross the ties to close the bag.
- 6. Make sure the bag is closed securely by inverting the bag to test the seal (no water leaks out).
- 7. Immediately place the Whirl-pak® bag into the cooler with ice and the blank. Optimal holding time for samples on ice or refrigeration is less than 6 hours, but not more than 24 hours.

BLANK DEC.13, 2022 9:10AM MARY MAYFLY

WHIRL-PAK





incubator.

BACTERIA MONITORIN PROTOCOL

Step 3: Plating your samples

Note: Turn incubator on before plating to ensure it will reach the correct incubation temperature of $35^{\circ}C \pm 1^{\circ}C$. Process the blank/control sample first (use 1 plate) and then follow with the stream sample (use 3 plates). **Use 1 pipette tip for your blank and 1 pipette tip per Whirl-pak® bag**. Plate the blank, remove and discard the tip, and pick up a new tip before plating your stream sample. Plates should reach room temperature before opening and plating (this only takes a few minutes).

- 1. Clean working area with disinfectant spray and let dry.
- 2. Put on personal protective gloves. NOTE: You should always wear these when handling and reading the plates.
- 3. Correctly label plates (1 Blank & 3 for your site sample), and lay them on a clean, flat surface. Plates should indicate stream name, site number, plate number, and the incubation start time and date.

11. Repeat: Plate two more samples for a total of three plates per stream site.

12. Leave plates undisturbed for one minute to allow the gel to solidify and then place them in the

- 4. Gently shake Whirl-pak® bag to ensure an even mix of sample.
- 5. Place the Whirl-pak® bag in a cup to keep from spilling and open the bag using the white tabs. *IMPORTANT! Do not touch the inside of the Whirl-pak® as this may contaminate your sample and alter the results.*
- 6. Carefully remove pipette tip from sterile container using the pipette. Don't touch the pipette tip with your hands or allow it to touch anything other than the water sample to avoid contamination.
- 7. Pipette 1 ml of the sample using the fixed-volume pipette: Push the pipette plunger down to the first "stop", submerge the tip into the sample, and release the plunger. This will load exactly 1 ml of water into the pipette tip. Always hold the pipette vertically while the tip is full of water.
- 8. Lift the clear top film of the 3M Petrifilm[™] plate by a corner and dispense the 1 ml of sample on the center of the circular plate,

the center of the circular plate, pushing the plunger all the way down to the second "stop". DO NOT let the pipette tip touch the plate.

- 9. Slowly roll the top film down onto the sample until the plate is completely covered to prevent trapping air bubbles. Do not touch the center of the plate.
- 10. If necessary, distribute the sample evenly using the 3M spreader (not included in all kits) or slightly tilting the plate back and forth. Tilting too much will cause the sample to pour out of the plate. If this happens, dispose of the plate and start a new one.

Lift the top film of the plate by a corner and release 1 ml of your sample onto the pink circle. Be careful not to touch the circle with the pipette tip or your hand.





Discard and resample if sediment is stirred up and collected in the sample bag.

Step 4:Incubating

- 1. Plan to turn on the incubator prior to plating to ensure it will be ready. Place the incubator lid on top.
- 2. Insert the digital max/min thermometer into the incubator.
- 3. Once the incubator is at $35^{\circ}C \pm 1^{\circ}C$, plate your samples, place the processed plates in the incubator, and reset the thermometer.
- 4. Incubate plates in a horizontal position, with the top film side up. Incubate for 24 hours \pm 1 hour at 35 \pm 1 degrees Celsius. If sampling more than one site, you can stack up to 20 plates in the incubator.
- 5. After 23-25 hours, remove plates (with gloves on) and count E. coli colonies.
- 6. Record the minimum and maximum temperatures that are displayed on the thermometer after incubating, as well as the time in/out of the incubator. If the incubation temperature has fluctuated outside of the 34-36°C range, the samples must be discarded.
- 7. Record all data on the Bacteria Data Form.
- 8. Dispose of plates: Open each plate, spray with an appropriate disinfectant, place them in a sealed zip lock bag, and discard in the trash.

Step 5: Cleanup & Disinfect

Properly disinfect your lab space, incubator, and cooler using 10% bleach disinfectant.

Reading the Results

When reading 3M PetrifilmTM plates, *E. coli* colonies appear blue and are closely associated with entrapped gas. General coliform colonies appear bright red and closely associated (approximately one colony diameter) with entrapped gas. *Remember that we are only concerned with counting the E. coli colonies in the medium* (*pink circle*), and we do not count colonies that appear on the foam barrier of the plate (or are more than 50%)

off the medium). Gas bubble patterns associated with gas producing colonies are shown at right. **Only count blue colonies that have a gas bubble!**

Bacteria growths on plates are counted as colony forming units per 100 milliliters of water sample (CFU/100 ml). Each 3M Petrifilm[™] plate holds 1ml of sample. Your final result for your sample is the average count of the three plates multiplied by 100 ml.

Getting "High" Bacteria Counts

SC AAS recommends that *E. coli* sampling be done monthly during normal flow conditions. Using guidance provided by the US EPA,

states have developed ambient standards for fecal coliform bacteria and/or *E. coli*. It is common to find high bacteria counts in urban areas, especially after rain events. *E. coli* counts (CFU/100 ml) that exceed 349 CFU/100 ml are above recreational standards and should be closely monitored, but when counts exceed the 1000 CFU/100 ml threshold, they warrant special action.

QUICK NOTE lerous to count (TNTC) and that have one

with gas-producing colonies.

Exception: You might encounter a plate with colonies that are too numerous to count (TNTC) and that have one or more of the following characteristics: 150 *E. coli* colonies or more, many gas bubbles, and deepening of the gel color from red to purple-blue. High concentrations of *E. coli* will cause the entire growth area to turn a deep purple-blue color. The plate will be filled with colonies with barely any empty space present. If this happens, please email a member of the State Team and resample as soon as possible.



BACTERIA MONITORII PROTOCOL

The following are examples of bacteria growth on 3M PetrifilmTM slides.

BACTERIA MONITORING PROTOCOL



BLANK





TOO NUMEROUS TO COUNT (TNTC)



4 CFU

STEP I. Count total <i>E.coli</i> colonies on all three of your stream sample plates and add them together.	Let's assume you counted 6, 7, and 8 colonies = 21 colonies
STEP II.* Find the average number of colonies. Take the total number of colonies and divide them by the number of plates used.	21 colonies / 3 plates = 7
STEP III.* Now, multiply the average number of colonies by 100. You have now determined the number of colony forming units per 100 ml of sample.	7 x 100 = FINAL COUNT 700 CFU/100 mL

*The SC AAS Database does this automatically.

In the SC AAS Database, a result of 1000 CFU/100 ml or higher will trigger an email alert to the SC AAS State Team, SCDES, and the local municipality or county, if they have agreed to receive such alerts.

If you find a "high" bacteria count, it may be a one-time event or occurrence. This information is useful! Add it to the database and return to the site as soon as possible to take another bacteria sample. When you return, pay careful attention to anything out of the ordinary at the site. Look for the presence of animals and be alert for any unusual odors. Walk the banks again (if you have access and permission) to look for obvious sources of pollution and note past and current weather conditions. Be sure to wear gloves while sampling and wash your hands carefully afterwards.

If you continue to find counts above the 1000 CFU/100 ml threshold, a member of the SC AAS State Team may reach out to you for more information. You may also wish to alert your local watershed group or local agency about your monitoring efforts and the results so far. They may be able to work with you on determining possible sources of *E. coli* pollution.

Note: The petrifilm process is used for screening purposes only. This process is not comparable to the SC State Standard. SC Adopt-a-Stream is not designed to identify or fix pollution sources.

Follow these steps if you have detected E. coli counts above 1000 CFU/100 mL:

- 1. Enter the result in the SC AAS Database so that alerts can be sent to the SC AAS State Team.
- 2. Re-sample as soon as possible and enter results as a second sampling event.
- 3. If re-sample value still exceeds 1000 CFU/100 mL, the SC AAS State Team may reach out to you for additional information.

Source Tracking

One method for determining sources of *E. coli* is called Microbial Source Tracking (MST). MST is a collective group of methodologies to determine actual sources of fecal pollution in environmental samples, which can include domestic pets, cows, deer, geese, hogs, other wild animals, birds, and humans. MST methods can be used in the design and implementation of Best Management Practices (BMPs) to reduce fecal loading in water.

Across the nation, the accuracy of molecular (genotypic) and biochemical (phenotypic) MST methods is being further evaluated by universities, agencies, and cities. Quantitative polymerase chain reaction (qPCR) is being looked upon most positively as having potential for standardization; this MST method recognizes target strands of DNA in a sample and amplifies them to a quantifiable level.

References

Frenzel, S.A., C.S. Couvillion (2002) Fecal-indicator bacteria in streams along a gradient of residential development. Journal of the American Water Resources Association. 38:265-273.

BACTERIA MONITORING PROTOCOL





Powerful and Secure Data Sharing

The SC AAS Database is a secure, searchable, publicly accessible resource that allows for the sharing of water quality data between certified volunteers, local governments, organizations, and your community. Data can only be added by certified volunteers, but can be viewed by anyone, with volunteers' personal information hidden. The SC AAS Database is where your data really has power to affect change!

The Database can be accessed on your computer, smartphone, or tablet. The steps outlined in this section will help you get started and set up to begin sharing data. You can also visit *www.scadoptastream.org* to find additional resources if you have questions!

Alerts

The SC AAS Database sends alerts to the SC AAS State Team and any city/county officials or organizations who would like to know about potential threats to local water quality. An automated email is distributed when the following results are recorded and saved in the database:

- Site needs organized cleanup.
- Bacteria results exceed 1000 CFU/100ml.
- Water Odors other than "natural" are present.
- "Other" is selected for Water Color.
- Water temperature is greater than 43 degrees Celsius.
- Dissolved oxygen is < 3 mg/L.

If you are interested in receiving these alerts for your area, please contact the SC AAS State Team.



The SC Adopt-a-Stream Database homepage, located at scaas.app.clemson.edu.

Using the SC Adopt-a-Stream Database

Step 1: Register Your Account and Login

Once certified, SC AAS Trainers add participants to the SC AAS Database with email address, certification(s), date, and county. The database will generate an email to all newly certified volunteers with a prompt to complete your registration, which includes entering contact information and creating a password. Your email address becomes your unique username. Once your account is set up, you can login to the database at **scaas.app.clemson.edu** by clicking "Login" in the top right corner. When you're logged in, you will see your email address in the top right corner instead of the word "Login." Now you can get started with joining a Group, adding your Site, and posting a Sampling Event!

Step 2: Add or Join a Group

Whether you join SC AAS as an individual or with others, for safety's sake, monitoring should always be completed by more than one person. Working amongst a Group also adds credibility to the data being recorded and allows you to discuss observations, draw conclusions together about what's happening at your monitoring site, and invite more people to become stewards of our watersheds. So talk to your family, neighbors, and friends about creating your sampling team!

The SC AAS Database requires you to join or create a new Group to be able to post data. Non-certified volunteers can always join you for monitoring, but remember that only certified volunteers can enter data, and only certified volunteers are listed as part of your Group in the database. Navigate to the Groups page by clicking the link at the top of the page, as shown below. You can then either: 1. Use the Search bar to find an existing Group, then just click the plus sign to the right of the name to join, or 2. Create a new Group by selecting the purple "Add Group" button and completing the form.

SC Adopt-A-Stream	m Sampling Events Groups Sites Certif	ications SC AAS Home		0	youremail@website.com
SC Adopt-A-Stream / Groups		 Navigate to the Groups page 			
Search for anything	Search Add Group	Ехро	t < 2		>
T <u>Search</u> for an existin	g Group or <u>create a new one</u>				
Group ID ↓≞_↓≓	Name ↓≞_↓≓	County ↓≞_↓≓	Registration Date ↓≞_↓≓		Options
32	CU-SWU Joint AAS Project	Pickens	05/20/2014		© View +
33	Upstate Master Naturalist Association	Pickens	09/04/2016		© View +
34	Crain K&G	Pickens	11/06/2016		© View +

USING THE SC AAS

ATABASE

The pop-up window shown here will appear when you select the "Add Group" button. Enter a Group name (this can be fun and creative), county, registration date, description, any existing monitoring Sites, and Group members.

- County: Start typing your county in the text box and a list will appear with similarly named counties in the U.S. Be sure to select the correct county and state.
- Monitored Sites: If your Site has already been created in the Database, begin typing the Site name or Site ID in the text box. Find and select your Site from the list. When entering data, you will only have the option to add data for sites that are monitored by your Group(s).

	×	Add Group	
	Name		
Add Group			< 1
	County		
e ta_t≓			ration Date ↓≞_↓≓
1	Registration Date		2/31/1969
m UF			06/14/2017
ok5	Description		6/14/2017
Discovery			6/19/2017
r Foundation	Monitored Sites	li	06/19/2017
r Falls	Search		6/28/2017
nty Stormwater	No sites have been selected.		06/30/2017
/ Mitchell	Members		8/06/2017
le Midlands	Search		16/30/2017
pyright © 2022 C	No members have been selected.		echnical assistance

• Members: To add Group members, start typing the name or username of a certified volunteer, then select them from the list. Be sure all Group members are added before saving. **Don't forget to add yourself!**

When you're finished, click the purple "Add Group" button in the bottom of the window!

Step 3: Add Your Monitoring Site

Adding your monitoring Site to your Group, or "adopting" it, will then allow you to post data for that Site. You may be adopting an existing Site that is no longer monitored by a former volunteer, in which case

×	Add Site
Waterbody Name	🦉 Map Satellite
Eighteenmile Creek	a contraction of the second seco
Waterbody Type	Allegan
Freshwaters ~	400
Watershed	
03060101 - Seneca ~	
Latitude	Nettles Dog Park
34.675064205574984	es Pan
Longitude	Google Creat Keyboard shortcuts Map data 62022 Terms of Use Report a map error
-82.78471909021611	
County	
Pickens	
City	
Clemson	

you can just add it to your Group in the previous step. However, many volunteers choose to adopt new stream sites that need to be added to the database and map.

Navigate to the "Sites" page by clicking the button at the top of the SC AAS Database homepage. Click the purple "Add Site" button. A pop-up window will appear to enter Site details, including waterbody name, waterbody type, watershed, latitude, longitude, description, and your Group name.

- Waterbody Name: If you aren't sure of your stream's name, refer to the SC Adopt-a-Stream Atlas (p. 54). If it doesn't have a name, first make sure it fits the criteria for site selection described on p. 29; if it does, you can choose a name. The Waterbody Name will become the first part of your Site ID (ex: Rocky Fork becomes RF-####), so it's best to keep this simple.
- Watershed: To find the **8-digit HUC watershed** in which your site is located, follow the instructions on p. 54 using the SC Adopt-a-Stream Atlas.
- Latitude and Longitude: You can determine your Site's latitude and longitude in a few ways:
 - 1. Record your coordinates in degrees using a GPS or mobile device while at your Site, then type them in to the form.

- 2. Find your Sites coordinates using the SC Adopt-a-Stream Atlas.
- 3. Create your Site using a mobile device while at your Site, with your device location turned on. The map in this pop-up window will zoom to your location, then you can simply click on the map to drop a pin. Your latitude and longitude will be added to the form.
- 4. Use the +/- buttons to zoom in/out, and click and drag to pan around to find your Site location on the map. (Note: If your device location is off, the map will default to the middle of the ocean.)

County, City, and State will automatically be completed once your Site is selected.

After completing the form, click the purple "Add" button in the bottom of the window. You should then see a green banner saying, "Successfully created a site with ID ____" at the top of the page.

Step 4: Add a Sampling Event

Once your account, Group, and Site are set up, you can begin adding sampling data!

Navigate to the "Sampling Events" page and click the purple "Add Sampling Event" button. Work through the tabs labeled with blue font (shown in the image below), making sure to only complete the sections for Freshwater sites. Here are a few notes to keep in mind:

- Site: Only sites that have been adopted by your Group will be available in this list. Be sure you are adding data for the correct Site before saving.
- Participant List: Add all Certified Volunteers who participated in this sampling event.
- Sampling Time: Be sure to set this to the correct time and date of your sampling event.
- Data entry sections will primarily mirror your data sheet. Upload photos in the "Observations" tab.
- Saving: Use "Save as Draft" if you need to return to edit later, such as to add *E. coli* data the next day. "Save and Submit" will finalize your data entry.

SC Adopt-A-Stream	Data Entry											
Site Information	Weather	Observations	Chemical	Bacteria	Macroinvertebrate	es Fre	shwater Stream Habitat	Saltwater Ha	bitat	Save as Draft	Save and	Submit
Don't see your site	? Make sure	the groups you ar	e in are setup t	to manage th	e site you need to in	put data f	or.					
Select a Site												
Site												
1MC-0237 - 12 M	lile Creek (S	ampling upstream	of the old iro	.)								
Site ID: 1MC-0237			County: Picke	ns		City: C	entral		State	South Carolina		
Watershed: Senec	а		Waterbody Typ	pe: Freshwa	ters	Latitude: 34.763 Longitude: -82.793						
Monitoring Groups			Participant Li	st		Sampling Time and Distance						
No groups monit	or this site.		Search by r	name		Event Date and Sample Time Furthest Distance Traveled (one-way)				ay)		
							12/20/2022 9:17 AM		0			miles

Additional Resources

If you encounter errors or technical issues while using the SC AAS Database, please use the "Need technical assistance?" link at the bottom of the page. A number of resources are also available to help you navigate the Database and use it responsibly. Visit <u>www.scadoptastream.org</u> to find:

- 1. Webinars Find recorded Database webinars on the "Resources & Materials" page of our website. Upcoming webinars will be announced in the SC AAS E-News and included in the Events Calendar.
- 2. Database FAQ Detailed instructions and responses to frequently asked questions can be found on the "Database FAQ" page:
- 3. Contact Information If you have further questions, reach out to a member of the State Team or your Trainer for help. Find contact information on the "Trainers & Staff" page.



Using the SC Adopt-a-Stream Atlas

Getting Started

The SCDES Adopt-a-Stream Stream Atlas (<u>https://gis.dhec.sc.gov/scaas/</u>) is a user-friendly tool to help you identify a sampling site or find important details about a site you've already selected to adopt. You can zoom to your location or type an address into the search bar to explore potential sites near you. Existing SC AAS volunteer monitoring sites are indicated by green markers and SCDES Water Quality Stations are indicated by red markers (turn on these stations in the "Layer List"). Water bodies are shown using blue shapes and lines. The closer you zoom in, the more detailed the map will become, allowing you to find smaller water bodies.

After identifying potential sites, do an in-person check to ensure they are safely accessible, wadable streams that flow year-round and are deep enough to get a sediment-free sample. Once you've picked a site, click that location on the map to view site details you will need when adding your site to the SC AAS Database. Your site will be added to the Atlas after you have adopted it in the SC AAS Database. Note that the map is updated monthly, so there will be lag time in displaying new sites.

Using the Application

You can find detailed instructions for using the Atlas by clicking the "Info" icon in the upper right-hand corner of the webpage. Here are a few key tips to get started:

- You can change the background to a satellite view by selecting "Imagery" in the "Basemap Gallery".
- Turn layers on and off using the "Layer List".
- Hover over icons to identify the tools or use "Tool Labels" linked in the left-hand collapsible sidebar.
- Click anywhere on the map to view details for that location. After choosing a monitoring site, click the map in the exact place you plan to sample. A pop-up will display the Waterbody Name, Watershed Name, and Watershed Number (8-digit HUC). Latitude and Longitude will be displayed in the lower left-hand corner of the screen. With this information, you can head to the SC AAS Database to create your site.
- Access the SC AAS Database from the Atlas by clicking the "Database" link on the blue bar at the top of the page.



APPENDIX 1: SC AAS DATA FORMS

Stream Habitat Survey: For Rocky and Muddy Bottom Streams (circle one)

Group

_ Stream name or Site ID _____

_ Investigators _

Date

Stream habitat will be 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.



Aquatic Vegetation diagrams courtesy of Houghton Lake Improvement Board

Take two photographs, looking upstream and downstream, capturing banks and riparian zone on both sides.

Total first side

Page 2 of 3



Page 3 of 3

Please submit all data promptly to the SC Adopt-a-Stream www.scadoptastream.org

	SC Adop	t-a-Stream	FRESHWA	TER Data Fo	rm	
		Distance Traveled		Travel Time		Sit
Site iD		(miles, one-way)		(minutes, one-way)		te II
Monitoring		Sampling Time		Certified Participants		nfor
Group		(minutes)		(first and last name)		rma
Time		Date (mm/dd/yyyy)				tion

Rainfall	Inches	Weather	🗆 Sunny 🗆 Partly Cloudy 🗆 Intermittent Rain 🗆 Steady Rain	Wea
(in last 24 hours)	**REQUIRED** www.cocorahs.org	(present conditions)	**DO NOT SAMPLE during unsafe conditions or after rain events**	ther

	🗆 Dry 🗆 Flood (Over Banks) 🗆 High 🗆 Low	Water Color	🗆 No Color 🗆 Brov	vn/Muddy 🗆 Green 🗆 Milky/White		
vvater Level	🗆 Normal 🗆 Stagnant/Still	(use clear container)	\Box Tannic \Box Other:			
Matan Sunface	🗆 Clear 🗆 Oily Sheen 🗆 Algae 🗆 Foam	Watar Odar	🗆 Natural/None 🗆 G	asoline 🗆 Sewage 🗆 Fishy 🗆 Chlorine		
water Surface	□ Other:	water Odor	□ Other:			
	🗆 Site is clean 🗆 I cleaned site					
Trash Cleanup	Needs organized cleanup	water Clarity		Tent L Cloudy/Somewhat Turbid		
	Concern of illegal dumping?	(sediment?)	□ Opaque/ Turbid			
	Alerts are generated when unus	ual colors, odors, or ille	egal dumping are selecte	ed		
Llamanda	🗆 Steep Bank 🗆 Trash 🗆 Fast Current	Destavia Courses	🗆 Dog 🗆 Goose 🗆 Livestock 🗆 Human			
Hazaros	□ Other:	Bacteria Sources	□ Other:			
Convitu	🗆 Drug Abuse 🗆 Vagrancy 🗆 Animals	Fish Movement	Incised Culver	t 🗆 Perched Culvert 🗆 Low Flow		
Security	□ Other:	Barriers 🛛 Dam 🗆 Ot		her:		
Reach	Active Channel Width: ft	If outfall/nine is pres	If outfall /ning is present is it flowing offer			
Dimensions	Bank Full Width: ft	2 days of dry weather?		🗆 Yes 🗆 No 🗆 N/A		
(optional)	Depth to Water ft	S days of dry weather:				
Photos	🗆 Upstream 🗆 Downstream 🗆 Extra					
110100	**Email additional photos to scaas@des.sc.gov as a .JPEG file**					

Parameter	Test 1	Test 2	Units	Duplicate Precision	Reminders:	
Air		N1/A	Calaina (9C)	Nama	Take air temperature BEFORE water temperature. Stand in the	
Temperature		N/A	Ceisius (°C)	None	shade. Hold thermometer from the top or use a string to hang.	
Water		N1/A	Calaina (8C)	Nama	Read thermometer while in the water or quickly after removing.	
Temperature		N/A	Ceisius (°C)	None	Take reading in shade. Check thermometer for air bubbles.	
			Standard units 0.25		Use a white background behind colorimeter to read result. Check	Ω
рн					against both scales. Do not wear hats or sunglasses.	hem
Dissolved					Take water samples from exact same place. Check the bottles and	lica
Oxygen			mg/L or ppm	0.6	syringes for air bubbles. Add drops slowly, one at a time.	I/Pł
Collinity		N1/A		Nana	The refractometer must be calibrated using distilled water prior to	iysi
Salinity		N/A	ррі	None	taking each measurement. Take off hats and sunglasses.	<u>a</u>
		N/A			You may need to remix the tube. Do not wear hats or sunglasses	
Transparency			cm	None	and take measurement in the shade. Do not stir up sediment in the	
	□ >120cm?				stream.	

Custom Tests (optional) and any chemical changes since you last sampled?:

Reminder to repeat tests if not within DUPLICATE PRECISION.

D ()	2NA Detailfiles NAetherd Lies d2		DI L					
Bacteria	3M Petrifilm Method Used?		Blank:					
Method	🗆 Yes 🛛 No	Colony Count	Plate 1: Plate 2: Plate 3:					
Incubation	Start Finish	Bacteria Changes	**Only count BLUE colonies WITH gas bubbles. Results should b					
	Time:		discarded if the Blank/Control plate shows bacteria growth.**	Bao				
Date/Time	Date:			cteria				
		Commente						
Incubation	Max: Min:	Comments,						
Temperature	**Incubation must be 24 \pm 1 hours and at 35 \pm 1 °C.**	Concerns?						
Visit www.scadoptastream.org to view and enter data.								

Field Checklist:
Handbook
Pen
Nitrile gloves/eye protection
Thermometer
DO and pH kits
Waste/rinse bottles
Transparency tube
Camera
Whirl-Pak bag
Sink water sample on ice in cooler
Trash bag
Paper towels
Incubator set to 35 °C
Closed-toe shoes

Always check chemicals for expiration dates prior to sampling your freshwater stream.

Experiencing a fish kill, health hazard, or dangerous pollution event? Call the SCDES Emergency Hotline at 1-888-481-0125.

APPENDIX 2: KIT CHECKLIST

Freshwater Kit Supply

Dissolved Oxygen

- LaMotte Dissolved Oxygen Kit
- LaMotte glass test tube
- LaMotte direct read titrator 0-10 ppm
- LaMotte sample bottle

Chemical Refills

Full Kit Refill

- Manganous Sulfate Solution (4167G) Alkaline Potassium Iodide-Azide (7166G)
 - Sulfuric Acid (6141WT-G)
 - Sodium Thiosulfate (4169-H)
 - Starch Indicator (4170WT-G)

pН

LaMotte Wide-range pH Water Test Kit

Chemical Refills

pH Wide range indicator



Bacteria

- 3m Petrifilm E. Coli plates (50)
- Digital max/min thermometer
- Still-air Incubator
- Pipet- Fixed-volume 1000 uL
- Sterile pipette tips
- Whirl-pak bags 4oz- Box of 500
- Cooler bag
- Ice pack

Turbidity Tube

Turbidity Tube, 120 cm

Other Equipment

- 500-1000mL Polyethylene waste bottle
- Wash bottle, 500 mL
- Nitrile gloves
- Thermometer
- Backpack
- Paper towels
- Sharpie
- Dry-erase marker
- Pen
- Printed Data sheets, MSDS, Instructions

APPENDIX 3: GLOSSARY

GLOSSARY OF WATER QUALITY TERMS

Acid rain. Rain with a pH of 4.5 or less.

Aerobic. Life or processes that depend on the presence of oxygen.

Aggrading stream reach. Deposition is greater than erosion within the stream reach.

Algae. Green plants that occur as microscopic forms suspended in water (phytoplankton) and as unicellular or filamentous forms attached to rocks and other substrates. These plants lack roots, stems, flowers, and leaves, live mainly in water, and use the sun as an energy source.

Algal bloom. A sudden increase in the abundance of suspended (planktonic) algae, especially at or near the water surface, producing a green appearance to the water. Excess nutrient can cause an algal bloom.

Anaerobic. Refers to life or processes that occur in the absence of oxygen.

Anaerobic decomposition. The breakdown of organic material without oxygen.

Anoxia. A condition of no oxygen in the water. Often occurs near the bottom of eutrophic, stratified lakes in summer and under ice in winter.

Aquatic community. All the groups of plants and animals occupying a common body of water.

Bacteria. Microscopic, single-celled organisms.

Bank. The portion of the stream channel that restricts the movement of water out of the channel during times of normal water depth. This area is characterized as being the exposed areas on either side of the stream above water level.

Baseline. A level or concentration that is the norm.

Baseflow. That portion of stream flow originating from groundwater discharging into the stream.

Basin. Another word for a watershed.

Benthic. Describes all things associated with the bottom, or sediments, of a stream.

Biodiversity. Biological diversity in an environment as indicated by the numbers of different species of plants and animals.

Bioengineering: This process uses living plants to restore stream banks, which in turn can control erosion, sedimentation, and flooding.

Biotic index. A numerical value that describes the biological integrity of aquatic communities for a waterbody.

Brook. A natural, fresh-water stream that is smaller than a river.

Canopy cover. Overhanging vegetation that provides shade to a stream.

Channelization. The straightening of streams by eliminating the meanders or bends. A channelized stream resembles a ditch with few or no meanders.

Conductivity. A measure of a material's ability to conduct electricity. Reported in microsiemens per liter for water (or other liquids).

Creek. A natural, fresh-water stream that is smaller than a river. This term is sometimes used specifically for small streams in coastal areas.

Cultural Eutrophication. Accelerated enrichment of waters due to human activities. Excess nutrients from agricultural runoff, sewage, and other sources allow waters to support a higher amount of plant and animal matter than they would naturally.

Decomposer. An organism that feeds on and breaks down dead plant or animal matter, thus making organic nutrients available to the ecosystem.

Degrading stream reach. A stream reach where erosion is greater than deposition.

Deposition. A natural process in which sediment (sand, clay, gravel) falls out of the water, wind, or ice that carries it. In a stream, this process builds up stream banks – the opposite of erosion. Also called sedimentation.

Discharge, Flow. A measure of how much water passes a given point in a given time (m³/s).

Discharge permits. The maximum amount of a pollutant that an entity is permitted to release into a waterbody.

Dissolved oxygen (DO). The amount of oxygen dissolved in water. Higher amounts of oxygen can be dissolved in colder waters than in warmer waters. Dissolved oxygen is necessary to support fish and other aquatic organisms.

Diversity. A large variety of organisms.

Ecology. The study of relationships among living and nonliving things

Ecoregion. Large area within which local ecosystems reoccur in a more or less predictable pattern. Ecoregions provide a spatial framework for ecosystem assessment, research, inventory, monitoring, and management.

Ecosystem. A community of animals, plants, and microorganisms interacting within the physical and chemical environment.

Embeddedness. The degree that larger particles (boulders or gravel) in a stream are surrounded or covered by fine sediment.

Emergent vegetation. Plants living along the edges (or banks) of a stream that are rooted in sediment but grow above the water's surface.

Ephemeral stream. A stream that flows during the wet season and is dry in the dry season; see Intermittent stream.

Erosion. The wearing down and removal of soil, rock fragments, and bedrock through the action of running water, wind, moving ice, and gravitational creep (or mass movement).

E. coli (*Escherichia coli*). A bacterium of the intestines of warm-blooded organisms, including humans, that is used as an indicator of water pollution for disease-causing organisms.

Eutrophication. A gradual increase in the productivity of a lake ecosystem due to enrichment with plant nutrients, leading to changes in the biological community as well as physical and chemical changes. This is a natural process, but can be greatly accelerated by humans (see Cultural eutrophication).

Fecal coliform bacteria. The portion of the coliform group that is present in the gut or feces of warmblooded organisms. The presence of fecal coliform bacteria in water is an indication of pollution and potential human health problems.

Floodplain. An area on both sides of a stream where flood waters spread out during high flow. The surface may appear dry for most of the year, but it is generally occupied by plants adapted to wet soils. Plants and trees in floodplains also filter pollutants and sediment.

Geography. Study of land (what it looks like, what it's used for, etc.), the things living there, the people (who they are and what they do), the interactions among these things, and their locations.

Geology. The study of the earth's history, the materials that make up the earth, and the processes that act on the earth.

Groundwater. Water found beneath the earth's surface.

Habitat. The place where a plant or animal lives that has all of the conditions necessary to support its life and reproduction.

Habitat diversity. The range of habitats within a region.

Hydrogeology. The effect geology has on water quality and stream morphology.

Hydrologic unit code (HUC). A description of watersheds that indicates size and location of particular watersheds; a watershed address.

Hydrologic cycle. The continuous movement of water among the oceans, air, and the earth in the form of precipitation, percolation, evapotranspiration, and stream discharge.

Hydrology. The science of how water flows on top of, and below, the earth's surface.

Hypoxia. A condition of low dissolved oxygen levels in a waterbody that can result from the decay of plants and algae.

Impervious. Water cannot pass through; waterproof.

Intermittent stream. A stream that flows when there is adequate precipitation and is dry when there's not. The stream does not flow continuously.

Invasive Species. A species of plant or animal that is not native to a given ecosystem but whose presence might cause environmental harm to the system or harm to human health. Invasive species often thrive in new habitats because they have no natural predators in the new ecosystem to keep them in check.

Lake. A large body of water that has water all year long.

Leaf litter. Plants and plant parts that have recently fallen and are partially or not at all decomposed.

Leaf pack. Any cluster or gathering of leaves and organic debris on the edges of streams or washed up on the upstream side of large rocks, fallen trees, or logs in the stream.

Left bank. When facing downstream, the bank to your left.

Losing stream. A stream that loses flow to the groundwater system.

MPN – is the acronym used for Most Probable Number, and represents a method for estimating quantitative results of organism growth in sample medium.

Macroinvertebrates. A spineless animal visible without the use of a magnifying glass.

Benthic macroinvertebrates, which live in the bottom of streams and wetlands, are good indicators of water quality because they live in the same area most of their lives and differ in their sensitivity to pollution. Which macroinvertebrates you find – or don't find – in a stream indicates the pollution level of the water. Benthic macroinvertebrates include aquatic insects (such as dragonfly and damselfly larvae) and crustaceans (such as crayfish, snails, and clams).

Meander. A bend in a stream.

Nonpoint source pollution. A type of pollution whose source is not readily identifiable as any one particular point, such as pollution caused by runoff from streets, agricultural land, construction sites, and parking lots. Polluted runoff and pollution sources not discharged from a single point.

Nutrient. Any of a group of elements necessary for the growth of living organisms, such as nitrogen and phosphorus. Excessive supplies of phosphorus or nitrogen, however, may enhance plant growth in surface waters.

Nutrient enrichment. Elevated levels of nitrogen and/ or phosphorus in a waterbody that result in nuisance growths of algae or other aquatic plants.

Organic matter. Plant and animal material.

Pathogen. An organism capable of causing disease.

Pathogenic. Capable of causing disease.

Perennial stream. Stream that flows nearly all year long.

Pervious. Allows water to pass through.

Pesticides. Any substance or mixture of substances intended for preventing, destroying, repelling, or mitigating any pest. Though often misunderstood to refer only to insecticides, the term also applies to herbicides, fungicides, and various other substances used to control pests.

pH. A measure of acidity or alkalinity on a scale of 0 to 14. A pH of 7 is neutral, less than 7 is acidic, and greater than 7 is alkaline (basic).

Photosynthesis. The process by which green plants produce oxygen from sunlight, water, and carbon dioxide.

Physical weathering. Erosion caused by mechanical forces (e.g., water expanding as it freezes and breaking apart rocks).

Phytoplankton. Algae that are microscopic and suspended in water.

Point source pollution. Pollutants originating from a "point" source, such as a pipe, vent, or culvert. **Point source contamination.** Contamination stemming from a single, isolated source, such as a drainpipe or an underground storage tank.

Pollution. An undesirable change in the environment, usually the introduction of abnormally high concentrations of hazardous or detrimental substances, such as nutrients or sediment. The presence of any substance that harms the environment.

Pollution-sensitive organisms. Organisms that cannot withstand the addition of pollution to their aquatic environment.

Pollution-tolerant organisms. Organisms that can withstand polluted environments.

Pond. Body of water that has water in it year-round but that is smaller than a lake.

Pool. That portion of a stream that is deep and slow moving, often following a riffle.

Reagents. A mixture or substance for use in chemical analysis or other reactions.

Respiration. Oxygen consumption in living organisms.

Riffle. That portion of a stream that is shallow and fast moving. An area of the stream where shallow water flows swiftly over completely or partially submerged rocks or other debris.

Right bank. When facing downstream, the bank to your right.

Riparian zone. An area adjacent to and along a watercourse that is often vegetated and constitutes a buffer zone between nearby lands and the watercourse. The natural plant community adjacent to a stream.

Riprap. Any material (such as concrete blocks, rocks, car tires, or log pilings) that may have been used to stabilize a stream from erosion.

Row cropping. A method of farming used in the production of corn and beans.

Run. A stream habitat type characterized as having a moderate current, medium depth, and smooth water surface.

Runoff. Water from rain, snowmelt, or irrigation that flows over the ground surface and runs into a waterbody.

Sanitary sewer. A pipe that carries food and human wastewater to a municipal sewer or septic system.

Secchi disc. A device used to measure the depth of light penetration in water.

Sediment. Eroded soil particles (soil, sand, and minerals) transported by water.

Sedimentation. The process by which soil particles (sediment) enter a water body, settle to the bottom, and accumulate. The addition of soils to lakes or streams.

Silt. Fine particles of soil and minerals formed from erosion of rock fragments.

Siltation. The process of silt settling out of water and being deposited as sediment.

Soil. Soil is a mixture of solids (minerals and organic matter), liquid, and gases that occurs on a land surface Slope. Change in elevation over a given distance.

Stable stream reach. A stream segment where sediment deposition is equal to erosion (i.e., no net gain or loss of sediment within the reach). It maintains its dimensions, pattern, and profile through time.

Stormwater sewer. A pipe that transports stormwater and meltwater runoff from roads and parking lots to streams and lakes. Stormwater sewers rarely lead to any type of treatment facility; the water is piped directly to streams and lakes. **Stream.** This term is used to describe any natural body of running water that moves over the Earth's surface in a channel or bed. Rivers, creeks, and brooks are all considered "streams."

Streambank. The sides of the stream that contain the flow, except during floods.

Streambed. The bottom of a stream where the substrate and sediments lie.

Stream depth. A measurement of the depth of a stream from the water's surface to the streambed.

Stream energy. Erosion potential of a stream.

Stream flow. The amount of water moving in a stream in a given amount of time.

Stream morphology. The shape of a stream. Stream order. Stream classification system. Stream reach. A specified length of stream.

Stream transect. An imaginary line drawn from water's edge to water's edge perpendicular to stream flow.

Substrate. The surface upon which an organism lives or is attached. The material making up the bottom of the streambed.

Suspended load. Sediment that is transported in suspension.

Thermal pollution. The raising of water temperatures by artificial means that interferes with the functioning of aquatic ecosystems. Sources of thermal pollution include removal of trees along streams, introduction of cooling water from power plants or other industrial facilities, and runoff from hot paved surfaces.

Tolerant species. An organism that can exist in the presence of a certain degree of pollution.

Topographic map. A map representing the surface features of a particular area. Features illustrated include streams, lakes, roads, cities, and elevation.

Topography. What the surface of the earth looks like.

Total coliform bacteria. A group of bacteria that is used as an indicator of drinking water quality. The presence of total coliform bacteria indicates the possible presence of disease-causing bacteria.

Transparency. The measure of water clarity. Transparency is affected by the amount of material suspended in water (i.e., sediment, algae, and plankton).

Turbidity. The presence of sediment in water, making it unclear, murky, or opaque.

Velocity. The speed at which water moves.

Water cycle. The continuous circulation of water in systems throughout the earth involving condensation, precipitation, runoff, evaporation, and transpiration.

Water ecology. The study of aquatic environments and the relationships among the living and nonliving things associated with those environments.

Water quality. The condition of the water with regard to the presence or absence of pollution.

Water Table. The top of the underground area that is filled with groundwater.

Watershed. A region or area of land that drains into a body of water such as a lake, river, or stream.

Wetland. Shallow body of water that may not have water in it year round.

APPENDIX 4: BEFORE YOU GO...

NEXT STEPS

Thank you for becoming a certified SC Adopt-a-Stream Volunteer! Now that you've completed your training workshop, you can get started collecting and sharing stream data!

- Activate Your Profile: Your Trainer will add you to the SC Adopt-a-Stream Database using the email address you provided when registering/signing in. Keep an eye on your inbox! You'll receive an email from SCAdoptAStream-L@lists.clemson.edu inviting you to activate your profile. Once you're logged in, you'll be able to get started with adding yourself to a group and adopting a site!
- Add your Group in the SC AAS Database: Groups are important for volunteer safety and active sharing of knowledge and result in long-term, supportive monitoring. Joining a Group is the first step to getting set up to share event data in the Database. Find instructions to create a Group or join an existing on in Chapter 7.
- Add your Site in the SC AAS Database: If you already know where you want to monitor, you can
 add your Site in the Database following the instructions in Chapter 7. If you need help finding an
 accessible stream site to adopt, feel free to contact your Trainer or a member of the State Team
 for guidance!
- Find a Kit: You can choose to purchase your own kit (via the <u>USC Upstate Marketplace</u>) or borrow one from one of the many organizations and partners around the state that offer kits for loan. Visit <u>www.scadoptastream.org</u> and check out the Kit Loan and Hub maps to find an available kit near you. Hubs are also locations where you can find support and connect with other local volunteers.
- **Start Monitoring:** Now you can head to the field and conduct your first monitoring event! If needed, you can find additional resources and training videos at <u>www.scadoptastream.org</u> to help you feel confident and prepared to start sampling.
- **Recertify in One Year:** Volunteers must maintain an active certification in order to continue posting in the SC AAS Database. You can recertify by attending another workshop or by completing our online course in March, July, or November. Don't worry-- we'll email you with more information when your certification is close to expiring.

Thank you for joining the SC Adopt-a-Stream community and contributing your time and efforts toward the protection of South Carolina's waterways!

South Carolina Freshwater State Standards Cheat Sheet								
Water Temperature	рН	Dissolved Oxygen	Transparency	E. coli.				
No single value for state standards	Between 6.0 and 8.5	Must average 5mg/L and be no less than 4 mg/L	Not to decrease below 16.6 cm on the transparency tube (50 NTUs)	Single sample shall not exceed 349 CFU/100 mL SC AAS Standard: sample shall not exceed 1000 CFU/100 mL				
Trout waters: no single value for state standards	Trout waters: between 6.0 and 8.0	Trout waters: no less than 6 mg/L	Trout waters: not to decrease below 52.2 cm on the transparency tube (10 NTUs)	Trout waters: same as above				

These are the SC State Standards for most freshwater streams; however, site-specific regulations do exist. For more information and to determine if a site-specific regulation exists in your area, refer to SC DES Regulation 61-69, Classified Waters.





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