

Macroinvertebrate Monitoring Methods Manual

Created by the Alliance for Aquatic Resource Monitoring at Dickinson College
for monitors participating in the Chesapeake Monitoring Cooperative in
Pennsylvania & New York



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This Macroinvertebrate Monitoring Methods Manual outlines the Chesapeake Monitoring Cooperative's (CMC) Macroinvertebrate Monitoring Program and contains information specific to the program and to ALLARM monitors in New York and Pennsylvania. The details of the CMC Macroinvertebrate Monitoring Program can be found in the Quality Assurance Project Plan, developed in May 2016, by the Chesapeake Monitoring Cooperative.

The purpose of this manual is to ensure that the methods used to collect and identify macroinvertebrates are consistent among ALLARM monitors throughout the upper Chesapeake Bay Watershed. The procedures written in this manual serve a quality control mechanism for collecting data of known, documented quality.

The CMC project is currently funded by EPA through December 2021.

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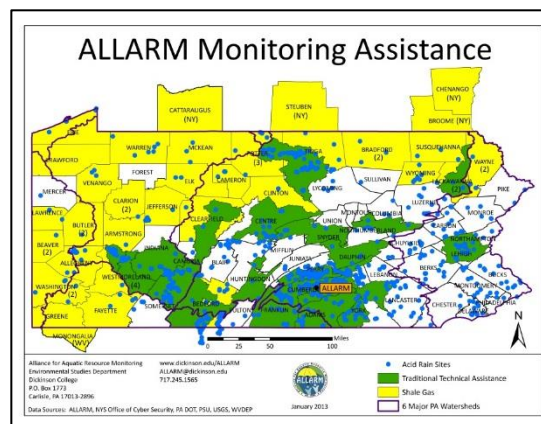
Background on ALLARM

The Alliance for Aquatic Resource Monitoring (ALLARM) is a nationally recognized technical and programmatic support center for community organizations interested in watershed assessment, protection, and restoration. ALLARM was founded in 1986, and is a project of Dickinson College. ALLARM's program goals are to:

- 1) Enhance local action for the protection and restoration of Pennsylvania watersheds by empowering communities with scientific knowledge and tools to implement watershed assessments;
- 2) Provide Dickinson College students with opportunities to participate in community-based participatory research thereby enhancing the quality of undergraduate science education; and
- 3) Be the leader in volunteer monitoring in Pennsylvania and a national model for college-community partnerships.

ALLARM provides customized assistance to community groups through skill-building workshops, trainings, and outreach on the following topics:

- Study design
- Chemical monitoring
- Macroinvertebrate monitoring
- Visual assessment
- Shale gas monitoring
- Quality assurance/quality control
- Data analysis and interpretation
- Communication strategies
- Strategic planning
- Volunteer recruitment



Through the work of three full-time staff, a science advisor, and 15 Dickinson College students, ALLARM offers comprehensive services to enable groups to use critical scientific tools that can enhance the quality of their local environment and allow them to participate fully in community decision-making. For more information on ALLARM, please visit: www.dickinson.edu/ALLARM.

Chesapeake Monitoring Cooperative

The objective of the *Integrating Citizen-based and Nontraditional Monitoring into the Chesapeake Bay Program Partnership* ("Project") is to gather stream data collected by citizen and non-traditional monitoring groups within the Chesapeake Bay Watershed. While the Chesapeake Bay Program (CBP) has immediate access to agency (federal and state) data collected in the watershed, non-traditional data producers are scattered throughout the 64,000 square mile watershed, collecting information that would be useful to the CBP. The integration of these data into the CBP monitoring network will provide additional cost-effective data and information to support shared decision-making and adaptive management by the CBP partners focused on restoration of the Chesapeake Bay and its watershed.

The Project is managed by the Alliance for the Chesapeake Bay (ACB) in partnership with a project coordinator at the Izaak Walton League of America (IWLA), and includes project partners at Dickinson College's Alliance for Aquatic Resource Monitoring (ALLARM) and University of Maryland Center for Environmental Science Integration and Application Network (UMCES). These four organizations collaborate to run the Chesapeake Monitoring Cooperative (CMC) and the three CMC monitoring programs: 1) Non-Tidal Water Quality Monitoring Program, 2) Tidal Water Quality Monitoring Program, and 3) Macroinvertebrate Monitoring Program.

Creating a Monitoring Study Design

ALLARM works with CMC groups in New York and Pennsylvania to develop a monitoring study design that addresses the questions and concerns of the group. The decisions made during the study process include choosing:

- What to monitor – macroinvertebrates and/or water quality parameters
- How to monitor – methodology and equipment used
- Where to monitor – identify and prioritize monitoring sites
- When to monitor – timeframe and frequency
- How to use and communicate the results

The study design process ensures that a monitoring program is designed with clear research questions and ways to answer those questions, based on how the data will be used. The final written document provides information on all aspects of the group's monitoring program.

Potential questions, goals, data uses of the CMC Macroinvertebrate Monitoring Program are summarized in the table below.

Questions	Goals	Results
<ul style="list-style-type: none">• What is the overall health of the stream?• Are there areas of the watershed that need additional assessment and/or restoration?	<ul style="list-style-type: none">• Measure stream health• Identify hotspots• Identify sites suitable for restoration projects	Data can be analyzed and used by monitors, groups, communities, stakeholders, government, and the CPB to: <ul style="list-style-type: none">• answer program questions• educate community and increase awareness of stream health issues• create a baseline of current watershed conditions to compare to if/when changes in the watershed occur• inform watershed management strategies at local and regional levels

CMC Macroinvertebrate Monitoring Program

The CMC Macroinvertebrate Monitoring Program aims to collect macroinvertebrate data throughout the Chesapeake Bay Watershed and store it in the CMC Database for use by the Chesapeake Bay Program, states, local government, and monitoring groups.

Each CMC group has a customized study design that addresses specific local questions. Although group questions and goals vary throughout the watershed, all monitors will follow the same guidelines to ensure that comparable data are being collected throughout the Chesapeake Bay Watershed. The minimum requirements to participate in the CMC Macroinvertebrate Monitoring Program include:

1. Create a written study design.
2. Attend CMC training workshops and follow-up meetings.
3. Become certified and maintain biennial certification.
4. Document your site location(s) using GPS and confirm that the site(s) is easy to access.
5. Follow the procedures included in this *CMC Macroinvertebrate Monitoring Methods Manual* as it applies to your study design – sample collection, identification, equipment maintenance, quality assurance & quality control, and data management.

CMC groups residing in the upper part of the Chesapeake Bay Watershed (PA & NY) are trained by ALLARM and follow the Environmental Alliance for Senior Involvement (EASI) Macroinvertebrate Monitoring Protocol. Monitors in the lower watershed are trained by IWLA and follow the VA Save our Streams Macroinvertebrate Monitoring Protocol.

Introduction to Macroinvertebrates

Macroinvertebrates are animals that are large enough to see without magnification and do not have a backbone. Examples include crustaceans, insect larvae, and worms. They play critical roles in stream ecosystems, occupying numerous feeding levels and being the primary group of organisms that can process leaves and detritus, which are the major source of energy in streams. They are also an important food source for other stream organisms, such as vertebrates and other large macroinvertebrates.

Macroinvertebrates spend much or all of their life in a stream, and the number and diversity of organisms living in the stream are influenced by: 1) the quality of the water, and 2) the availability and condition of in-stream habitat. Macroinvertebrate populations are threatened by chemical and thermal pollution, sedimentation, and habitat loss. Different types of macroinvertebrates tolerate different levels of pollution and stream conditions, which make them good indicators of stream health. Healthy streams have an abundant and diverse macroinvertebrate community, with a predominance of pollution-sensitive species.

Macroinvertebrates are most abundant in riffles and runs (areas of fast moving water), where fine sediment does not accumulate and where oxygen is plentiful. Some types are also found in areas around banks or in-stream structures that provide protection, such as large rocks and logs. The protocol outlined below focuses on collecting macroinvertebrates from these two kinds of habitats.

EASI Macroinvertebrate Monitoring Protocol

ALLARM trains monitors to follow the Environmental Alliance for Senior Involvement (EASI) Macroinvertebrate Monitoring Protocol, a volunteer-friendly protocol adapted from the [EPA Volunteer Stream Monitoring Methods Manual](#). This protocol is used throughout the upper Chesapeake Bay Watershed by volunteers looking to learn more about the health of their local streams. Macroinvertebrates are collected and identified to the Order level, then counted and placed into one of three pollution categories (sensitive, somewhat sensitive, or tolerant). The number and diversity of organisms found in the pollution tolerance categories are inserted into the metric to classify a stream reach as having good, fair, or poor water quality. It is suggested that macroinvertebrates be monitored twice a year – once in the spring and once in the fall, in order to account for seasonal changes within the stream and the lifecycle of some organisms.

Monitoring Macroinvertebrates

Safety Considerations

You should always consider safety first, and should never monitor when you feel ill, during inclement weather (especially snowy/icy conditions), or under high-flow conditions. Take caution when entering and exiting the stream and wear waders or close-toed shoes. It is good practice to have a first aid kit for cuts and scrapes.

Choosing a Method

The EASI Protocol contains two different sampling methods, and you will first need to choose which method (rocky bottom streams vs. muddy bottom streams) is most suitable for your monitoring site, based on the stream's hydrology and streambed:

Stream Reach Characteristic	Rocky Bottom Stream	Muddy Bottom Stream
Hydrology		
Riffles (Shallow, fast-moving water)	X	
Runs (Deep, fast-moving water)	X	
Pools (Deep, slow-moving water)		X
Streambed		
Boulders (> 10" in diameter)	X	
Cobbles (2.6 – 10" in diameter)	X	
Gravel (~0.2 – 2.5" in diameter)	X	
Sand, Silt, or Mud (< 0.2" in diameter)		X

Site Visit Preparation

Before you visit your monitoring site, gather all of the equipment and supplies needed to collect and identify macroinvertebrates, and make sure they are clean and in good condition. These items may include:

Monitoring Binder	Site Set-Up
<ul style="list-style-type: none">• <i>Macroinvertebrate Methods Manual</i>• Field Data Sheet• Identification & Scoring Sheet	<ul style="list-style-type: none">• Tape measure• Flagging to identify 100-foot reach• GPS unit
Macroinvertebrate Collection	Macroinvertebrate Identification
<ul style="list-style-type: none">• Fishing license• Bucket• Waders or close-toed shoes• Sample bottle• First aid kit• Rocky Bottom Streams:<ul style="list-style-type: none">○ Kick net (1 m x 1 m, 500 µm mesh)• Muddy Bottom Streams:<ul style="list-style-type: none">○ D-net (1 ft)○ Gloves○ Wash bottle○ Sieve	<ul style="list-style-type: none">• Sorting tray – dish pan, ice cube trays, containers• Sorting utensils – spoons, tweezers, paint brushes• Magnification – hand lens, microscope• Identification key(s) and resources• Camera• Pen• Calculator

Macroinvertebrate Collection: Rocky Bottom Streams

For rocky bottom streams, a kick net is used to disturb three, 1-meter x 1-meter areas located in riffles/runs to collect a representative sample of the macroinvertebrate population. The collection procedure requires at least two team members – a “net holder” and a “kicker”.

Identify the stream reach

When you arrive at the stream, fill out the Sampling Information section of your Field Data Sheet. Scan the area to identify a 100-foot stream reach to monitor, and choose three sites within the reach to sample. The three sites should be in areas of fast-moving water (riffles/runs) and, if possible, have different flow velocities and substrate sizes (gravel to cobble) in order to get a better representation of the in-stream habitat diversity. Use a tape measure and flagging to mark off the 100-foot reach you will be sampling. On your Field Data Sheet, draw a sketch of the stream reach you have chosen to monitor and record the GPS coordinates of the upstream and downstream endpoints. Label your three sites (#1 is downstream, #2 is midstream, and #3 is upstream).

The stream reach should be at least 50 feet upstream from any human-made structure, such as a dam or bridge.

Get into place

Enter the stream downstream of site #1 and carefully wade upstream to the site. The net holder should face upstream and place the kick net in the water, holding it at a 45-degree angle to the water's surface. This angle can be adjusted accordingly, depending on the water depth, as long as water **DOES NOT** flow over the top of the net, which could result in the loss of macroinvertebrates! The bottom of the net must be snug against the bottom of the streambed. This can be achieved by: 1) sewing a chain into the bottom of the kick net to weigh it down, or 2) using rocks *from the sampling area* to anchor the bottom of the net. If using rocks, rub them in front of the net underwater to remove any macroinvertebrates that may be clinging to them, before placing them on the bottom of the net.

Dislodge the macroinvertebrates

Once the kick net is in place, the kicker should stand 1 meter away from the net, facing downstream. First, pick up any large rocks in the 1-meter X 1-meter sampling area upstream of the kick net and rub them thoroughly underwater so that any macroinvertebrates clinging to the rocks will flow into the net. Be sure to check for case-building caddisflies, as they tend to stick securely to the rocks. Place the “clean” rocks outside of the kicking area. Next, start at the upstream end of the 1-meter X 1-meter kicking area, and begin disturbing and kicking up the streambed (\geq two inches in depth). Kick the entire 1-meter x 1-meter for approximately three minutes.

Remove the kick net from the stream

The kicker should remove any rocks used to anchor the net to the bottom of the streambed. Next, the net holder grasps the top handles and the kicker grabs the bottom handles. Together, lift the net out of the water using a scooping motion, and carefully exit the stream, staying downstream from site #2.

When removing the kick net from the stream, make sure water does not flow over the top or bottom of the net...otherwise you will lose some of the organisms that you just collected!

Remove the macroinvertebrates from the kick net

Immediately release any fish, amphibians, or reptiles that were captured in the net. If you collect crayfish, record how many you collected on your Identification & Scoring Sheet, and release them back into the stream. You can either pick the macroinvertebrates directly from the net or flush the contents collected into a bucket. If picking from the net directly, place it on a flat surface and transfer the macroinvertebrates to containers or sorting trays with enough water to cover all of the organisms while you collect the macroinvertebrates from sites #2 and #3.

Collect the second and third sample

Repeat the process for sites #2 (midstream) and #3 (upstream), and combine all of the organisms from the three sites. This will provide a better representation of the stream's overall macroinvertebrate community.

Macroinvertebrate Collection: Muddy Bottom Streams

For muddy bottom streams, 20 “jabs” with a D-frame net in different habitat areas are combined to get a representative sample of the macroinvertebrate population. This collection procedure requires only one monitor. There are typically four types of habitat found in muddy bottom streams:

1. Vegetated bank margins
2. Snags and logs
3. Aquatic vegetation beds
4. Decaying organic matter

Identify the sampling area

When you arrive at your monitoring site, fill out the Sampling Information section on your Field Data Sheet. Scan the area to identify a 100-foot stream reach to monitor. Use a tape measure and flagging to mark off the 100-foot reach you will be sampling. The sampling area should be at least 50 feet upstream from any human-made structure, such as a dam or bridge. On your Field Data Sheet, draw a sketch of the stream reach you have chosen to monitor and record the GPS coordinates of the upstream and downstream endpoints.

Determine the number of jabs for each habitat category

Identify the types of habitat present in the stream reach, and record them on your Field Data Sheet. Use the chart below to determine how to split up the 20 jabs, based on the number of habitats present:

	Vegetated bank margin	Snags and logs	Aquatic vegetation beds & decaying organic matter	Silt/sand/gravel substrate
4 habitats present & abundant	10 jabs	Combination of 10 jabs		
3 habitats present & abundant	Combination of 15 jabs			5 jabs
2 habitats present & abundant	Combination of 15 jabs			5 jabs
Some habitats abundant and others sparse OR if you cannot reach a habitat type due to unsafe conditions	Jab the sparse habitats as much as possible, then jab the remaining abundant habitats. (20 jabs total)			

Get into place

Enter the stream downstream from the 100-foot reach and fill your bucket and wash bottle with stream water.

Dislodge the macroinvertebrates

Use the following methods to collect the macroinvertebrates from each type of habitat present using the D-net:

- a. **Vegetated bank margin:** All jabs should be done underwater – jab vigorously, with an upward motion, brushing the net against the vegetation and roots along the bank.
- b. **Snags and logs:** Hold the net with one hand under the submerged wood. With the other hand (gloved), rub 1 sq. ft. of the snag or log. Scoop everything you dislodge into your net.
- c. **Aquatic vegetation beds and decaying organic matter:** All jabs should be done underwater – jab vigorously, with an upward motion, against or through the plant bed.
- d. **Silt/sand/gravel substrate:** Place the flat part of the net against the stream bottom and push it forward one foot upstream to dislodge ~3 inches of the streambed.

Record the number of jabs taken from each type of habitat on your Field Data Sheet as you move upstream.

Remove the macroinvertebrates from the D-net

Transfer the contents from each jab into the bucket filled with stream water. Hold the D-net over the bucket and use the wash bottle to help remove anything stuck to net. After completing 20 jabs, pour the contents from the bucket through a sieve to concentrate the debris and organisms collected. Sort through the sample and transfer the organisms (look for anything moving) to the sorting trays.

Macroinvertebrate Identification

Use the sorting supplies to divide the macroinvertebrates into groups of organisms – each group should have similar characteristics and appearance. Use a hand lens (if needed), the Identification & Sorting Sheet, and any other resources to identify the organisms to the Order level. Once all of the macroinvertebrates have been sorted and identified, count the number of organisms collected for each type of macroinvertebrate listed on the Identification & Scoring Sheet, and record the number in the box to the right of the macroinvertebrate.

Unknown Specimens

If you (or anyone else in your group) cannot identify an organism, follow the directions below:

1. Photograph the organism – take multiple shots, and include an object in the picture for scale.
2. Fill out the Unknown Specimen section of your Field Data Sheet.
3. E-mail your photographs, Field Data Sheet, and completed Identification & Scoring Sheet to ALLARM (ALLARM@dickinson.edu) for identification.

Water Quality Score Calculation

The macroinvertebrates are categorized into three groups on the Identification & Scoring Sheet based on their ability to tolerate pollution:

Tolerance Groups	Pollution Tolerance	Quality of Water Found
Group I	Sensitive	Good
Group II	Somewhat sensitive	Good or Fair
Group III	Tolerant	Good, Fair, or Poor

Assign and record an abundance code (R, C, or D) for each type of macroinvertebrate, based on the number of organisms you collected and recorded on your Identification & Scoring Sheet:

- Rare (R) 1 – 9 organisms
- Common (C) 10 – 99 organisms
- Dominant (D) ≥ 100 organisms

Once the abundance codes have been assigned, follow the instructions on the back of the Identification & Scoring Sheet to calculate the water quality score of the stream reach you sampled. Record the final water quality score on your Field Data Sheet as well as the amount of time you spent monitoring your stream reach.

Wrap-up

Return the macroinvertebrates to the stream and rinse all of your monitoring equipment. All materials must dry completely before storing them. Review your Field Data Sheet and Identification & Scoring Sheet to be sure all sections were filled out completely.

Data Management

Field Data Sheet and Identification & Scoring Sheet

The Field Data Sheet is used to document the sampling and site details and the monitoring results. The Identification & Scoring Sheet is used to record the number of macroinvertebrates you collect and calculate the water quality score for your monitoring site. After monitoring, place both sheets in your ALLARM Monitoring Binder until you are able to give them to ALLARM for archiving. ALLARM recommends taking a photocopy or picture of your sheets before handing over the original copies to ALLARM. ALLARM will hold onto them for at least seven years following the sampling date.

Database

The CMC maintains an online database for monitors to upload their monitoring results. After you have filled out your Identification & Scoring Sheet completely, enter the information on your sheet into the database (*directions to enter data into the project database will be provided by ALLARM once the database has been created and is ready for use*). If you are not comfortable using the database, you can ask someone else in your monitoring group or ALLARM to enter your results.

Quality Assurance & Quality Control

Quality assurance and quality control (QA/QC) are the backbone of any successful stream monitoring program. QA/QC procedures ensure that the data collected are useful and of known quality. The CMC Macroinvertebrate Monitoring Program implements a variety of QA/QC practices, summarized in the table below:

Practice	Components
Training & Certification	<ul style="list-style-type: none">• Attend ALLARM training workshops and follow-up meetings.• Become an ALLARM certified monitor and maintain biennial certification (successfully collect and identify a sample – observed and verified by ALLARM).
Documented Procedures	<ul style="list-style-type: none">• Written study design describing the monitoring goals and approved by ALLARM.• Site location(s) documented using GPS and verified by ALLARM.• Sampling methods followed as written in this manual.
Equipment Maintenance	<ul style="list-style-type: none">• Equipment and materials inspected each time before use.• Equipment cleaned and stored appropriately.
Internal Verification	<ul style="list-style-type: none">• At least two monitors must sort through and identify the macroinvertebrates collected from each sample.
External Verification	<ul style="list-style-type: none">• Unknown specimens are photographed for identification by ALLARM.• Data sheets and data entered into the CMC Database are spot checked by ALLARM (minimum of 25% of sampling events).

Certification

You will need to be certified to collect and submit data to the CMC Database, or at a minimum be supervised by a certified monitor during sample collection and identification. Certification includes collecting, sorting, and identifying an entire sample while being observed by ALLARM. You will need to identify 90% of the organisms correctly in order to become a certified monitor. To maintain certification, you will need to repeat this process once every two years.



Field Data Sheet

CMC Macroinvertebrate Monitoring Program

Sampling Information

Record the information about today's sampling event in the boxes below.

Monitor Name(s)				
Date		Time		
Stream Name		Site Name		
Weather Conditions	Clear	Partly Cloudy	Cloudy	Fog/Haze
Precipitation	None	Drizzle	Rain	

Site Information

Draw a diagram of the 100-foot stream reach you have chosen to monitor. Record the latitude and longitude coordinates of the upstream and downstream endpoints.

Upstream	Lat:	Long:
Downstream	Lat:	Long:

Muddy Bottom Streams

Record the types of habitat types present and abundant in the 100-foot stream reach and the number of jabs taken from each habitat type (20 jabs total).

Habitat Type	Present & Abundant?	Number of Jabs Taken
Vegetated bank margin		
Snags and logs		
Aquatic vegetation beds & decaying organic matter		
Silt/sand/gravel substrate		

Unknown Specimen

Record information about any organisms that you cannot identify. The more information you provide, the easier it will be for ALLARM to identify the organism(s).

Organism #1					
Does the organism have legs?	Yes	No	If yes, how many legs?	6	> 6
Does the organism have a tail?	Yes	No	Approximate size		
Description of organism					
Organism #2					
Does the organism have legs?	Yes	No	If yes, how many legs?	6	> 6
Does the organism have a tail?	Yes	No	Approximate size		
Description of organism					
Organism #3					
Does the organism have legs?	Yes	No	If yes, how many legs?	6	> 6
Does the organism have a tail?	Yes	No	Approximate size		
Description of organism					

Final Results







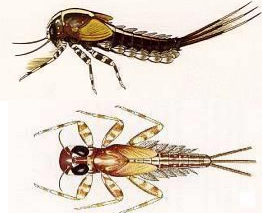





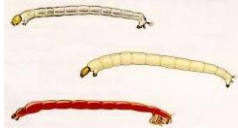




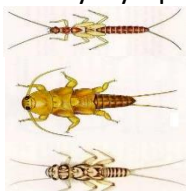




Water Quality Score (#)		Time Spent Monitoring	
Notes			



Identification & Scoring Sheet

CMC Macroinvertebrate Monitoring Program

Rare (R) = 1 – 9 organisms
 Common (C) = 10 – 99 organisms
 Dominant (D) = ≥ 100 organisms

Group I Sensitive	Count	Group II Somewhat Sensitive	Count	Group III Tolerant	Count
Water Penny Larvae 		Beetle Larvae 		Aquatic Worms 	
Hellgrammites 		Clams 		Blackfly Larvae 	
Mayfly Nymphs 		Cranefly Larvae 		Leeches 	
		Crayfish 			
Gilled Snails 		Damselfly Nymphs 		Midge Larvae 	
		Scuds 			
Riffle Beetles (adult) 		Sowbugs 		Snails 	
Stonefly Nymphs 		Fishflies 		Site Designation:	
		Alderflies 			
Non Net-Spinning Caddisfly Larvae 		Net-Spinning Caddisfly Larvae 		Team Members:	

Calculating the Water Quality Score

(From EPA Volunteer Monitoring Methods Manual)

To calculate the water quality score:

1. Record the number of R's, C's, and D's found for each Macroinvertebrate Group in box A.
2. Multiply each number (A) by the weight factor listed (B) and record the number in box C.
3. Add the three numbers in box C to get a total number for each Macroinvertebrate Group.
4. Add the totals for all three groups to get the water quality score for the stream reach monitored.

Group I Sensitive				Group II Somewhat Sensitive				Group III Tolerant			
A		B	C	A		B	C	A		B	C
# R's		x 5.0		# R's		x 3.2		# R's		x 1.2	
# C's		x 5.6		# C's		x 3.4		# C's		x 1.1	
# D's		x 5.3		# D's		x 3.0		# D's		x 1.0	
Group I Total =				Group II Total =				Group III Total =			

Water Quality Score = _____ + _____ + _____
 (Group I Total) (Group II Total) (Group III Total)

Water Quality Score = _____

<u>Water Quality Scores</u>	
> 40	Good water quality
20 – 40	Fair water quality
< 20	Poor water quality



Rocky Bottom Procedure (Summary)

CMC Macroinvertebrate Monitoring Program

A rocky bottom stream contains riffles/runs and the streambed is composed of gravel, cobbles, and boulders. If the stream does not have riffles/runs and has a muddy, silty, or sandy streambed, use the **Muddy Bottom Stream Procedure**.

Equipment & Supplies:

Monitoring Binder	Site Set-Up
<ul style="list-style-type: none"> • <i>Macroinvertebrate Methods Manual</i> • Field Data Sheet • Identification & Scoring Sheet 	<ul style="list-style-type: none"> • Tape measure • Flagging to identify 100-foot reach • GPS unit
Macroinvertebrate Collection	Macroinvertebrate Identification
<ul style="list-style-type: none"> • Fishing license • Bucket • Waders or close-toed shoes • Sample bottle • First aid kit • Rocky Bottom Streams: <ul style="list-style-type: none"> ○ Kick net (1 m x 1 m, 500 μ mesh) 	<ul style="list-style-type: none"> • Sorting tray – dish pan, ice cube trays, containers • Sorting utensils – spoons, tweezers, paint brushes • Magnification – hand lens, microscope • Identification key(s) and resources • Camera • Pen • Calculator

Step #1: Identify the sampling area

1. Choose a location at least 50 feet upstream from any human-made structure (dam, bridge, etc.) and one that has riffles/runs.
2. Use a tape measure and flagging to mark off a 100-foot stream reach. Record the latitude and longitude coordinates of the upstream and downstream endpoints.
3. Identify three riffle/runs areas within the reach for sampling and draw a sketch of the stream reach on your Field Data Sheet. Label the three sites (#1 is downstream; #2 is midstream; #3 is upstream).

Step #2: Get into place

1. Enter the stream downstream from site #1.
2. Insert the kick net into the stream, facing upstream, at a 45° angle to the water surface.
3. Place the bottom of the net tightly against the stream bottom – use large rocks within the 1-meter x 1-meter sampling area (rub them for macros first) or a chain in the net to help weigh down the net.
4. Once the net is in place, do not allow any water to flow over the net.

Step #3: Dislodge the macroinvertebrates

1. Pick up large rocks inside the 1-meter x 1-meter sampling area and scrape any macroinvertebrates clinging to the rocks into the net. Place the “clean” rocks outside of the sampling area.
2. Starting furthest away from the net, kick $\geq 2''$ of the streambed with your feet, working your way towards the net. Disturb the sediment in the 1-meter x 1-meter sampling area for ~ 3 minutes.

Step #4: Remove the kick net from the stream

Remove any rocks used to anchor the net. Together, use a scooping motion to remove the net out of the water, being VERY careful not to let any water to flow over the net.

Step #5: Remove the macroinvertebrates from the kick net

1. Return any fish, amphibians, and reptiles back to the stream.
2. Record the number of crayfish collected and return them to the stream.
3. Flush the contents collected into a bucket or place the net on a flat surface to remove the macroinvertebrates by hand, transferring them into containers filled with water.

Step #6: Collect the second and third sample

Repeat steps #2 – 5 for sites #2 and #3 in the stream reach.

Step #7: Sort the macroinvertebrates

Use the sorting supplies to divide the macroinvertebrates into groups of organisms – each group should have similar characteristics and appearance.

Step #8: Identify the macroinvertebrates

1. Use the identification keys and resources to identify the organisms. Use magnification if needed.
2. Count and record the number of organisms collected for each type of macroinvertebrate.
3. Photograph any organisms that cannot be identified and fill out the Unknown Specimen section of the Field Data Sheet.

Step #9: Calculate the water quality score

1. The macroinvertebrates are categorized into three groups based on their ability to tolerate pollution:

Tolerance Groups	Pollution Tolerance	Quality of Water Found
Group I	Sensitive	Good
Group II	Somewhat sensitive	Good or Fair
Group III	Tolerant	Good, Fair, or Poor

Assign and record an abundance code for each type of macroinvertebrate, based on the number of organisms collected:

- Rare (R): 1 – 9 organisms found
 - Common (C): 10 – 99 organisms found
 - Dominant (D): ≥ 100 organisms found
2. Follow the instructions on the back of the Identification & Scoring Sheet to calculate the water quality score of the stream reach. Record the score and the amount of time you spent monitoring on the Field Data Sheet.

Step #10: Wrap-up

Return the macroinvertebrates to the stream and rinse all of your monitoring equipment. All materials must dry completely before storing them. Review your Field Data Sheet and Identification & Scoring Sheet to be sure all sections were filled out completely.



Muddy Bottom Procedure (Summary)

CMC Macroinvertebrate Monitoring Program

A muddy bottom stream has a muddy, silty, or sandy streambed and does not contain riffles/runs. If the stream has riffles/runs and a streambed composed of gravel, cobbles, and boulders, use the **Rocky Bottom Stream Procedure**.

Equipment & Supplies:

Monitoring Binder	Site Set-Up
<ul style="list-style-type: none"> • <i>Macroinvertebrate Methods Manual</i> • Field Data Sheet • Identification & Scoring Sheet 	<ul style="list-style-type: none"> • Tape measure • Flagging to identify 100-foot reach • GPS unit
Macroinvertebrate Collection	Macroinvertebrate Identification
<ul style="list-style-type: none"> • Fishing license • Bucket • Waders or close-toed shoes • Sample bottle • First aid kit • Muddy Bottom Streams: <ul style="list-style-type: none"> ○ D-net (1 ft) ○ Gloves ○ Wash bottle ○ Sieve 	<ul style="list-style-type: none"> • Sorting tray – dish pan, ice cube trays, containers • Sorting utensils – spoons, tweezers, paint brushes • Magnification – hand lens, microscope • Identification key(s) and resources • Camera • Pen • Calculator

Step #1: Identify the sampling area

1. Choose a location at least 50 feet upstream from any human-made structure (dam, bridge, etc.).
2. Use a tape measure and flagging to mark off a 100-foot stream reach. Record the latitude/longitude coordinates of the upstream and downstream endpoints and draw a sketch of the stream reach.

Step #2: Determine number of jabs for each habitat category

Identify and record the types of habitat present in the stream reach and determine the number of jabs to make in each type of habitat.

	Vegetated bank margin	Snags and logs	Aquatic vegetation beds & decaying organic matter	Silt/sand/gravel substrate
4 habitats present & abundant	10 jabs	Combination of 10 jabs		
3 habitats present & abundant	Combination of 15 jabs			5 jabs
2 habitats present & abundant	Combination of 15 jabs			5 jabs
Some habitats abundant and others sparse OR if you cannot reach a habitat type due to unsafe conditions	Jab the sparse habitats as much as possible, then jab the remaining abundant habitats. (20 jabs total)			

Step #3: Get into place

Enter the stream downstream from the 100-foot reach and fill the bucket and wash bottle with water.

Step #4: Dislodge the macroinvertebrates

1. Collect the macroinvertebrates from each type of habitat present using the D-net:
 - a. **Vegetated bank margin:** All jabs should be done underwater – jab vigorously, with an upward motion, brushing the net against the vegetation and roots along the bank.
 - b. **Snags and logs:** Hold the net with one hand under the submerged wood. With the other hand (gloved), rub 1 sq. ft. of the snag or log. Scoop everything you dislodge into your net.
 - c. **Aquatic vegetation beds and decaying organic matter:** All jabs should be done underwater – jab vigorously, with an upward motion, against or through the plant bed.
 - d. **Silt/sand/gravel substrate:** Place the flat part of the net against the stream bottom and push it forward one foot upstream to dislodge ~3 inches of the streambed.
2. Record the number of jabs taken from each type of habitat as you move upstream.

Step #5: Remove the macroinvertebrates from the D-net

1. Transfer the contents from each jab into the bucket filled with water.
2. After completing 20 jabs, pour the contents from the bucket through a sieve.
3. Sort through the sample and transfer the organisms (look for anything moving) to the sorting trays.

Step #6: Sort the macroinvertebrates

Use the sorting supplies to divide the macroinvertebrates into groups of organisms – each group should have similar characteristics and appearance.

Step #7: Identify the macroinvertebrates

1. Use the identification keys and resources to identify the organisms. Use magnification if needed.
2. Count and record the number of organisms collected for each type of macroinvertebrate.
3. Photograph any organisms that cannot be identified and record them on the Field Data Sheet.

Step #8: Calculate the water quality score

1. The macroinvertebrates are categorized into three groups based on their ability to tolerate pollution:

Tolerance Groups	Pollution Tolerance	Quality of Water Found
Group I	Sensitive	Good
Group II	Somewhat sensitive	Good or Fair
Group III	Tolerant	Good, Fair, or Poor

Record an abundance code (R, C, or D) for each macroinvertebrate, based on the number collected:

- Rare (R): 1 – 9 organisms
 - Common (C): 10 – 99 organisms
 - Dominant (D): ≥ 100 organisms
2. Follow the instructions on the Identification & Scoring Sheet to calculate the water quality score of the reach. Record the score and the amount of time you spent monitoring on the Field Data Sheet.

Step #9: Wrap-up

Return the macroinvertebrates to the stream and rinse all of your monitoring equipment. All materials must dry completely before storing them. Review your Field Data Sheet and Identification & Scoring Sheet to be sure all sections were filled out completely.