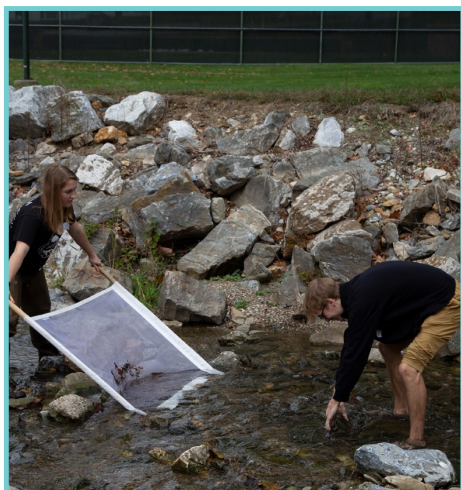


# Stream Team Macroinvertebrate Monitoring Manual

March 2020  
Version 2.1



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**Dickinson**

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# MACROINVERTEBRATE MONITORING MANUAL

This Macroinvertebrate Monitoring Methods Manual is designed for volunteer monitors participating in the Alliance for Aquatic Resource Monitoring (ALLARM) Stream Team program. The macroinvertebrate protocol is adapted from the Environmental Protection Agency's [Volunteer Stream Monitoring: A Methods Manual](#). The goal of this manual is to describe, in full detail, the directions and steps necessary for Stream Team volunteers to collect credible benthic macroinvertebrate data and calculate a water quality score. In order for data to be compliant with ALLARM's federally approved QAPP, we ask that Stream Teams follow testing instructions carefully. This manual has been adapted from the Chesapeake Monitoring Cooperative's (CMC) Macroinvertebrate Monitoring Program and contains information specific to ALLARM monitors in Pennsylvania and New York. Further details of the program can be found in the Quality Assurance Project Plan, developed in May 2016 (<http://j.mp/CMCmacroQAPP>).

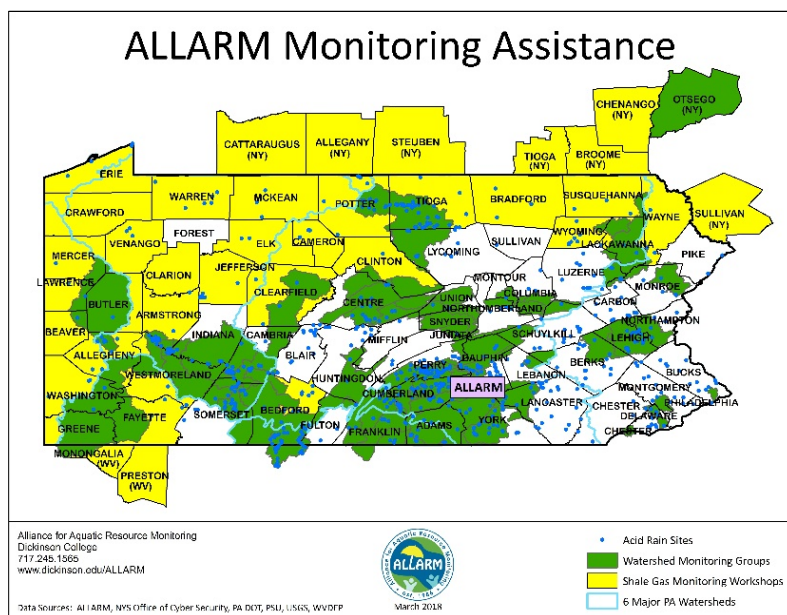
## ALLIANCE FOR AQUATIC RESOURCE MONITORING

The Alliance for Aquatic Resource Monitoring 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 in Carlisle, PA. ALLARM's mission is to empower communities with scientific tools to understand the health of their streams and participate in local decision-making.

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

- Study design
- Water quality
- Macroinvertebrate monitoring
- Visual assessment
- Quality assurance/quality control
- Data analysis and interpretation

Through the work of four full-time staff, a faculty science advisor, and 10-12 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 fully participate in community decision-making. For more information on ALLARM, please visit: [www.dickinson.edu/ALLARM](http://www.dickinson.edu/ALLARM).



## SUSQUEHANNA STREAM TEAM

The Susquehanna River is the largest tributary to the Chesapeake Bay, providing about 50% of its fresh water and draining close to 28,000 mi<sup>2</sup> of land. Improving the health of the Susquehanna River watershed is key to Pennsylvania and New York achieving Bay pollution reduction goals. Stream monitoring is a tool for assessing the health of the Susquehanna watershed and determining the

effectiveness of the Bay pollution reduction plan. To this end, ALLARM developed a protocol for chemical and biological stream monitoring to collect baseline data on Susquehanna tributaries. These data can contribute to local community decision making, as well as to regional data analysis through the Bay-wide central database ([Chesapeake Data Explorer](#)).

## ACKNOWLEDGEMENTS

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The ALLARM Susquehanna Stream Team volunteer monitoring program is funded through a number of programs, including the Campbell Foundation, in partnership with the Lower Susquehanna Riverkeeper Association. Funding support is also provided by the Chesapeake Monitoring Cooperative, a cooperative agreement with the Environmental Protection Agency's Chesapeake Bay Program; ACB CB96334901 – Citizen and Non-traditional Monitoring, as well as the PA Department of Environmental Protection funded Consortium for Scientific Assistance to Watersheds (C-SAW). ALLARM appreciates all of its funders and partners in this project, including local watershed associations, county conservation districts, Penn State extension offices, and all of the volunteer monitors. Thank you for your collaboration.



## INTRODUCTION TO MACROINVERTEBRATES

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Macroinvertebrates are animals that are large enough to see without magnification (macro) and do not have a backbone (invertebrates). 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.

## IMPORTANT CONSIDERATIONS

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This macroinvertebrate monitoring protocol is catch and release. However, because the process is extremely stressful to the organisms, not all the macroinvertebrates may survive. This is one of the trade-offs of biomonitoring; in order to get important data on what is living in the stream, it can cause harm to the individuals sampled. However, the percentage of insects we remove from the stream compared to those that remains is unlikely to harm the populations of aquatic organisms, only individuals. In scientific research using vertebrates, strict protocols are put in place to ensure that the benefit of the obtaining the data outweighs the harm done to the animals. Most invertebrates, including aquatic invertebrates, are exempt from this oversight. However, we still want to minimize the impact on aquatic life. General tips to limit impact to aquatic life from sampling include:

- choosing sites with banks that won't break down
- minimizing walking in a stream beyond the necessary sampling
- keeping containers with macroinvertebrates out of direct sunlight or from undergoing large temperature changes
- separating organisms into by type to avoid predator-prey interactions
- returning insects to the stream they were found

There are also legal considerations when working with live organisms. In the state of Pennsylvania, the collection of aquatic macroinvertebrates is considered fishing, which is regulated by the Fish and Boat Commission. In order to collect aquatic macroinvertebrates, a fishing license or educational permit is needed. During our workshops, you will be covered under our educational permit. However, you will need a fishing license for the day you sample. If you don't already have a fishing license or intend to get one, we recommend getting a [one-day fishing license](#) for the day you plan to do your macroinvertebrate monitoring. In addition to a fishing license, some stocked streams are off-limits for any type of fishing (including macroinvertebrates) in the months of March and early April. Please consult the Fish and Boat Commission's [webpage](#) for these dates.

## MACROINVERTEBRATE MONITORING PROTOCOL

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ALLARM trains monitors to follow a volunteer-friendly protocol adapted from the EPA's [Volunteer Stream Monitoring: A Methods Manual](#). 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.

### Site Safety Considerations

The number one rule for stream monitoring is safety first! Please check the weather and stream conditions before going out to the field. Always monitor in a team of at least 2 people. Take caution when entering and exiting the stream and wear waders or closed-toe shoes. It is good practice to have a first aid kit available to attend to cuts and scrapes, as well as a cell phone. Be aware of hazards in the area. You may be exposed to hazards such as insects, poisonous plants or animals, uneven terrain, and strenuous activity. It is not possible to list each risk associated with participation in this program.

In addition to being able to enter the water safely and legally, there are other factors for choosing a good macroinvertebrate collection site. It is important to determine whether your site is a feasible

location for this macroinvertebrate monitoring protocol. Preferable conditions include low water flow, abundant riffle zones, a wide bank, and more than a 50-foot distance upstream from man-made structures like dams or bridges. If your site does not fit these characteristics, you can look for a better site on your stream or join another team for the macroinvertebrate monitoring portion of the protocol.



Picture A is bad because the water is too high, moving too fast, and there is no accessible bank.  
 Picture B is good because the water is low and there is a wide, safe bank for access.

## 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"> <li>• <i>Macroinvertebrate Methods Manual</i></li> <li>• Field Data Sheet</li> <li>• Identification &amp; Scoring Sheet</li> </ul>	<ul style="list-style-type: none"> <li>• Tape measure</li> <li>• GPS unit or cell phone</li> </ul>
Macroinvertebrate Collection	Macroinvertebrate Identification
<ul style="list-style-type: none"> <li>• Fishing license</li> <li>• Bucket</li> <li>• Waders or close-toed shoes</li> <li>• Sample bottle</li> <li>• Rocky Bottom Streams:                             <ul style="list-style-type: none"> <li>○ Kick net (1 m x 1 m, 500 µm mesh)</li> </ul> </li> </ul>	<ul style="list-style-type: none"> <li>• Sorting tray – dish pan, ice cube trays, containers</li> <li>• Sorting utensils – spoons, tweezers, paint brushes</li> <li>• Magnification – hand lens, microscope</li> <li>• Identification key(s) and resources</li> <li>• Camera</li> <li>• Pen</li> <li>• Calculator</li> </ul>

## 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 assess and choose three areas within the reach to sample. The three areas should be in 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 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 assess and record the GPS coordinates of the upstream and downstream endpoints. Label the three areas (#1 is downstream, #2 is midstream, and #3 is upstream).



### Get into place

Enter the stream downstream of area #1 and carefully wade upstream to the area. 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 according to 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 from the sampling area, 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) as you slowly move towards the net. 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 of the poles and the kicker grabs the bottom of the poles. Together, lift the net out of the water using a scooping motion, and carefully exit the stream, staying downstream from area #2. Try to keep the net outstretched and horizontal as you exit the stream as opposed to folding the net in half.

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

### 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. Place the net on a flat surface like the stream bank and transfer all of the macroinvertebrates to containers or sorting trays filled with enough water to cover all of the organisms while you collect the macroinvertebrates from areas #2 and #3.

### Collect the second and third sample

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

## Macroinvertebrate Identification

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

### 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](mailto: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

The overall evaluation of the site is based on the relative abundance of organisms from each of the groups. 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

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. Macroinvertebrate data will be uploaded into the Chesapeake Data Explorer.

# APPENDIX



# Macroinvertebrate Field Data Sheet

## Sampling Information

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

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

## Site Information

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

Upstream	Latitude:	Longitude:
Downstream	Latitude:	Longitude:

## 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). Be sure to send along a photograph as well.

<b>Organism #1</b>					
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					
<b>Organism #2</b>					
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					
<b>Organism #3</b>					
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					







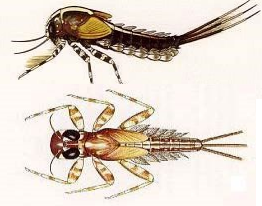





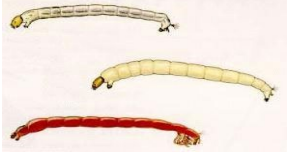




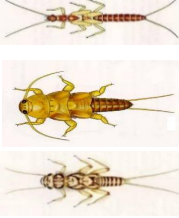




### Final Results

Water Quality Score (#)		Time Spent Monitoring	
Notes			



# Macroinvertebrate Identification & Scoring Sheet

- Count and record the number of organisms collected for each type of macroinvertebrate.
- Record an abundance code for each macroinvertebrate type, based on the number of organisms collected:
  - R** (rare) = 1 – 9 organisms
  - C** (common) = 10 – 99 organisms
  - D** (dominant) =  $\geq$  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 		Crane-fly Larvae 		Leeches 	
		Crayfish 			
Gilled Snails 		Damselfly Nymphs 		Midge Larvae 	
		Scuds 			
Riffle Beetles (adult) 		Sowbugs 		Snails 	
Stonefly Nymphs 		Fishflies 		Site Name/ID:	
		Alderflies 		Sampling Date:	
Non Net-Spinning Caddisfly Larvae 		Net-Spinning Caddisfly Larvae 		Monitor Names:	

# 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 result in box C.
3. Add the three numbers in box C to get a total value for each Macroinvertebrate Group.
4. Add the totals for all three Groups to get the water quality score for the stream reach sampled.

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	
<b>Group I Total =</b>			<b>Group II Total =</b>			<b>Group III Total =</b>		

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

Water Quality Score = \_\_\_\_\_

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



# Rocky Bottom Procedure (Summary)

## Stream Team 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, contact ALLARM to use the **Muddy Bottom Stream Procedure**.

### Equipment & Supplies:

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

### 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. 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):  $\geq 100$  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.