

Chapter VII

How to Collect, Process, and Identify Benthic Macroinvertebrate Samples

"Chemical measurements are like taking snapshots of the ecosystem, whereas biological measurements are like making a videotape."

Prof. Dr. David M. Rosenberg PhD

If you've tried to find methods for your program, you've probably discovered that there is a wide variety of methods out there. Further, the originators of each method are absolutely convinced (and will try to convince you) that their method is the "best." And they may be right . . . for their waters and for their purposes. The challenge for you is to figure out which method you will use to meet your program goals.



The answer may be simple: do what your state agency does, or what your colleagues are doing, or simply the easiest. But as we emphasized in chapter V, you should select a method that is appropriate for your goals, needs, resources and capabilities.

For this reason, we've opted to present a "menu" instead of a prescription.

IN THIS CHAPTER . . .

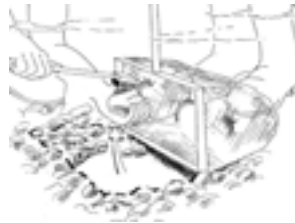
- 1) **Methods for Collecting and Preserving Samples**
- 2) **Lab Methods for Processing Samples**
- 3) **Lab Methods for Identifying Samples**
- 4) **Complete Field Methods**

1. Methods for Collecting and Preserving Samples

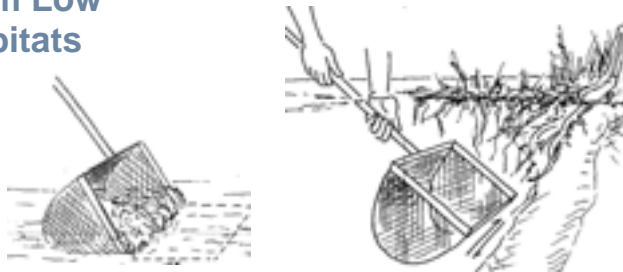
TYPES OF COLLECTION METHODS

There are 3 basic types covered in this chapter:

A. Collecting Samples from Riffles Using Nets



B. Collecting Samples from Low Gradient Habitats



C. Collecting Samples from Riffles or Deep Sites Using Artificial Substrates



1. Methods for Collecting and Preserving Samples

Within each of these types, there are several methods. Several features that differentiate these methods are described below.

Degree of Quantification

The type of collection device, which is driven partly by whether you need to collect qualitative, semi-quantitative, or quantitative samples, differentiates these methods. This has to do with the extent to which you can say how many critters are in your sample:

- * quantitative methods produce a count
- * semi-quantitative methods produce an extrapolated estimate
- * qualitative methods produce a characterization (e.g. abundant, rare, dominant)

Quantification is described more fully in Chapter V as one of the main things to consider when selecting a method.

of Replicate Samples

Critters do not disperse themselves equally throughout the stream segment. Replicate sampling is a quality control step that assures that your samples will be representative of the stream segment. A replicate sample is another sample collected from the same segment as your sample. Basically, the more spots you hit, the more likely you are to collect all the types of critters that are there in the proportions that reflect the whole segment.

Simple versus composite samples

A simple sample is one collection from one spot in the target habitat. A composite sample is a sample that is made up of 2 or more collections at different spots in the same habitat.

A. Collecting Samples from Riffles Using Nets

The methods described on the following pages are designed to collect samples from riffles - fast moving water, relatively shallow (<18"), and flowing over cobbles (rocks 2-10") in diameter using some form of net.

Riffles are great habitat for aquatic invertebrates for several reasons:

- * The water tends to be well-oxygenated
- * There are lots of attachment surfaces
- * There are lots of little niches among the cobbles, behind larger rocks, etc.
- * Food is carried through the riffle and can be eaten in a variety of ingenious ways designed to remove the food from the current.
- * Many of the most pollution sensitive critters live there.

For all these reasons, riffles tend to have the most diverse community of invertebrates, just waiting to pass along information.



This method is the simplest in this manual. It does not rigorously standardize the level of effort per sample and it does not require the amount of work per sample that method A2 does. Because of this, the results can't be expressed as the number of organisms per sample and the number of organisms collected can not be compared among the sites sampled.

The following steps should be carried out by at least two people: one to hold the net (the "holder") and the other to dislodge critters (the "sampler").



THE METHOD AT A GLANCE

Habitat Sampled	Riffle bottom
Sampling Device	Metal frame net or sieve
Level of Effort	Not standardized
Quantitative?	No
# Samples	1 composite sample from 1 fast spot and 1 slow spot in the riffle

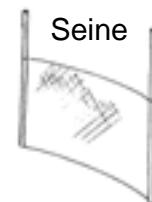
Note: This method can be used with seine nets and D-nets, but the procedures on the following pages will need to be adjusted slightly.

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- * Collection Field Sheets
- * Collection Net (500-600 mm mesh): 18" X 8" rectangular metal frame OR 500-micron mesh Surber Sampler
- * Heavy-Duty Arm-length Gloves
- * 5-Gallon Bucket and Sieve (#30 mesh)
- * Soft, Nylon Bristle Brushes
- * Forceps
- * Wide Mouth Sample Containers (at least 1 liter): sealable plastic bottles or heavy-duty Zip-lock Bags - 1 for each replicate
- * 90% or 70% de-natured Ethyl Alcohol
- * Labeling Tape and Pencils

2) Follow directions to the first site and find the riffle you will be sampling.



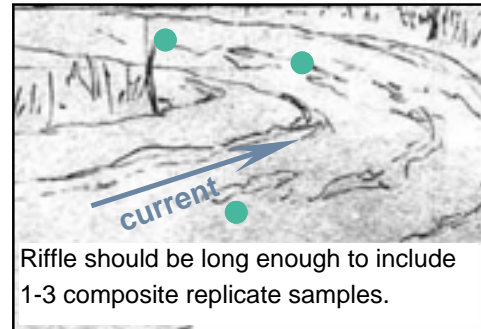
METHOD 1.A.1 NETS IN RIFFLES

COLLECTING A QUALITATIVE SAMPLE USING A NET

On-Site Prep List

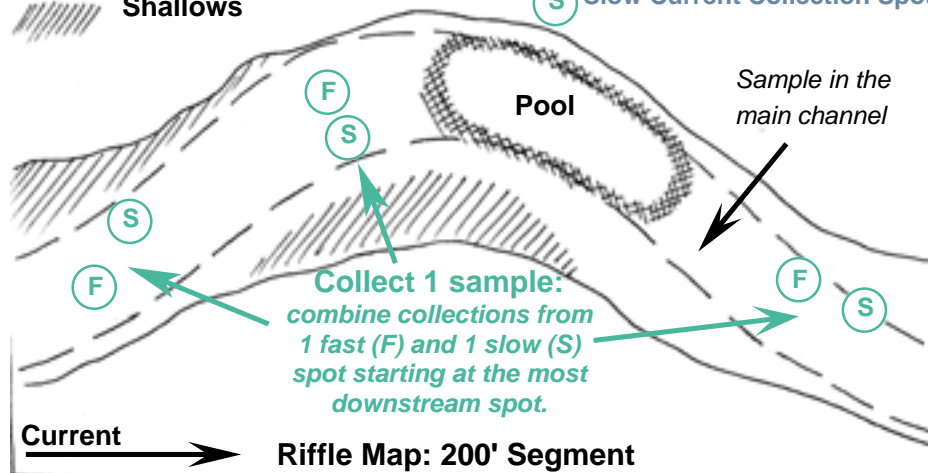
- 1) Fill in the top half of the *Collection Field Sheet*.
- 2) Measure a 200' segment that contains the riffle habitat and draw this segment on the back of the field sheet.
- 3) Identify your collection spots.
- 4) Enter the stream at the downstream side of the first spot and wade into position.

Possible Sample Locations



Tip:

Avoid shallow areas that might be dry at base (low) flow.



Tip:

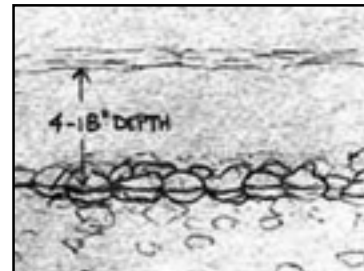
Avoid eddys behind rocks or your leg - they create an upstream flow that will allow your critters to escape.



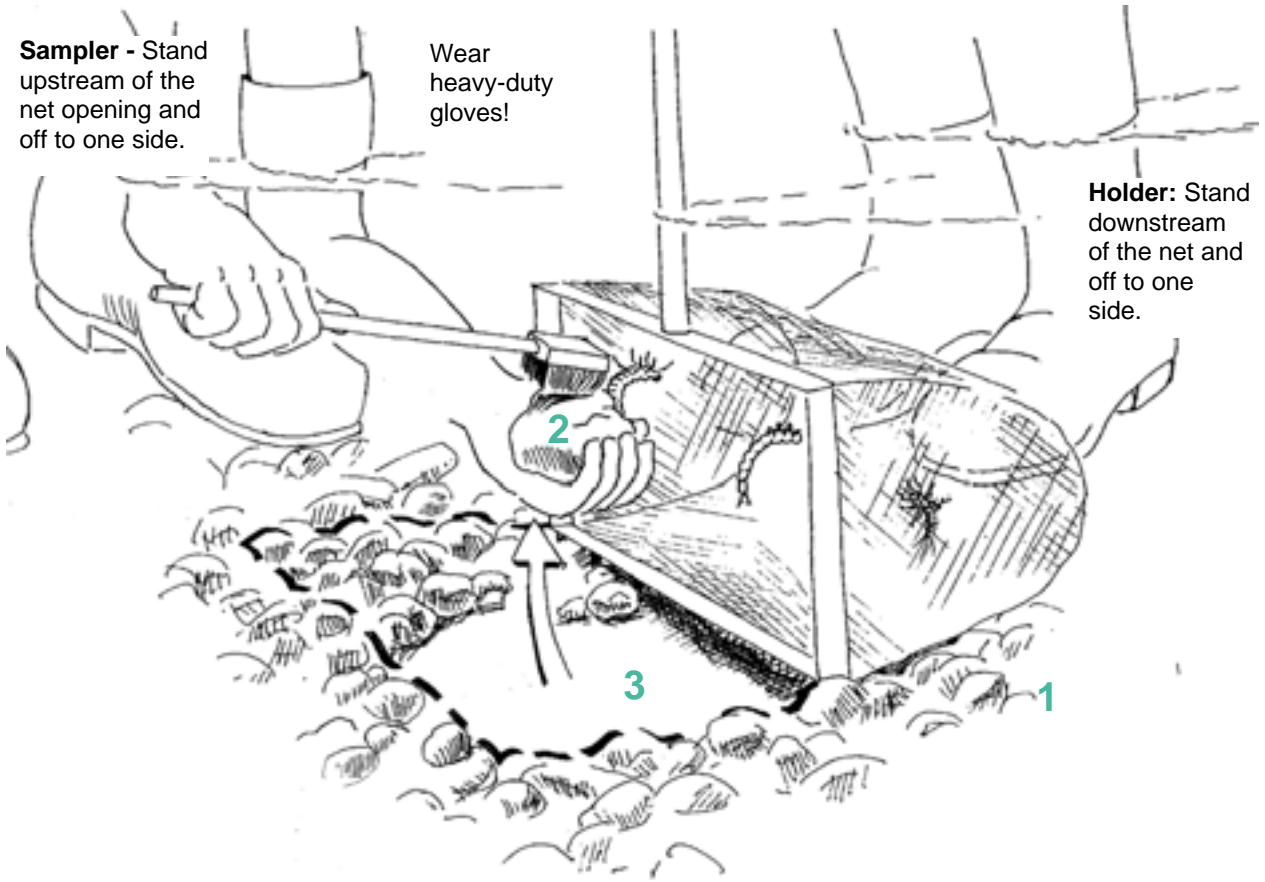
A bit too large . . .



Ideal cobble =
2-10"



Sample cobble bottoms, no greater than 4 - 18" deep

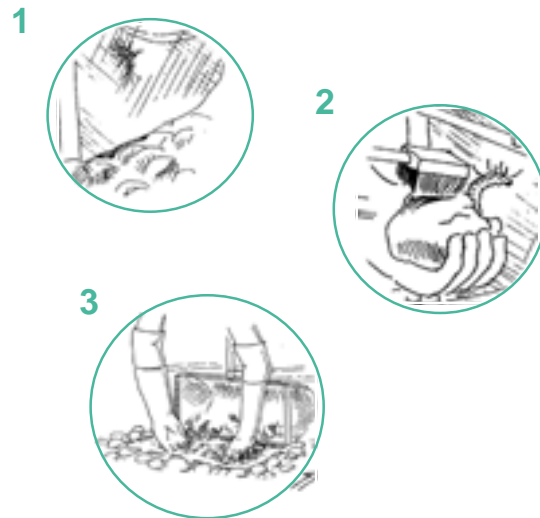


Step 1: Place the net on the bottom so that it creates a tight seal.

Step 2: Dislodge critters from the work area by lifting each cobble and rubbing or brushing it off toward the net. After cleaning, set each cobble aside out of the work area.

Step 3: Get the burrowers by gently digging into the soft bottom (if any) beneath the cobbles you removed with your hands or a stake.

Step 4: Lift the net out of the water and proceed to the next spot to collect the rest of the sample.



METHOD 1.A.1
NETS IN RIFFLES

COLLECTING A QUALITATIVE SAMPLE USING A NET

Start here after you've collected each sample (steps 1 - 4).

Is the sample loaded with sand and gravel? If so, consider "swirling" it. See page VII-23 for instructions

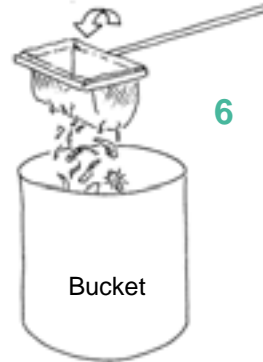


A preserved sample

Step 5: Inspect the sample - set the vertebrates free!



Step 6: Empty the contents of the net into a bucket.



Step 7: Pick the critters off of the large debris.



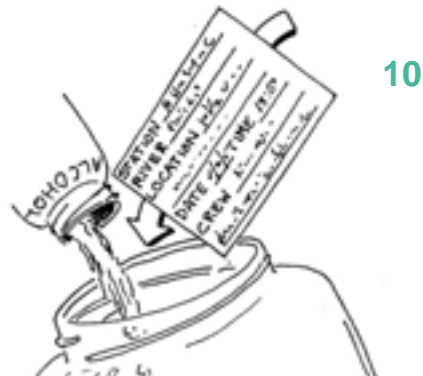
Step 8: Transfer the sample from the bucket to the sample container.



Step 9: Pick all remaining critters out of the net.



Step 10: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it. The label should contain the site #, replicate #, date, samplers initials, stream etc..



COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET

METHOD 1.A.2 NETS IN RIFFLES

This method is a rigorous semi-quantitative method that produces representative samples. Because the level of effort is standardized to an approximate area of stream bottom (0.55 m), you will be able to estimate the density of organisms per given area of stream bottom, which is useful in comparing the communities among all your sites. Finally, it allows you to use other data analysis tools to interpret your results.

The following steps should be carried out by at least two people: one to hold the net (the “holder”) and the other to dislodge critters (the “sampler”). Another person can do a more thorough cleaning of the cobbles (the “cleaner”), fill out the collection field sheet, and assist with sample processing and preservation.



THE METHOD AT A GLANCE

Habitat Sampled	Riffle bottom
Sampling Device	18" X 8" rectangular metal frame net w/ 500-600 mm mesh
Level of Effort	Standardized by area
Quantitative?	Semi: area specified but not delineated by the sampling device: 0.55 square meters sampled per replicate.
# Samples	1 – 3 composite replicates: Each replicate is a composite of sampling 2 fast and 2 slow areas for a total of 12 collection spots.

Note: This method can be used with seine nets and D-nets, but the procedures on these pages will need to be adjusted slightly.

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- * Collection Field Sheets
- * Collection Net (500-600 mm mesh): 18" X 8" rectangular metal frame OR
- * 18" X 8" rectangular metal frame net w/ 500-600 mm mesh
- * Heavy-Duty Arm-length Gloves
- * 5-Gallon Bucket (2) and Sieve (#30 mesh)
- * Soft, Nylon Bristle Brushes
- * Forceps
- * Wide Mouth Sample Containers: sealable bottles or heavy-duty Zip-lock Bags - 1 for each replicate
- * 90% or 70% de-natured Ethyl Alcohol
- * Labeling Tape and Pencils

2) Follow directions to the first site and find the riffle you will be sampling.



Not drawn to scale

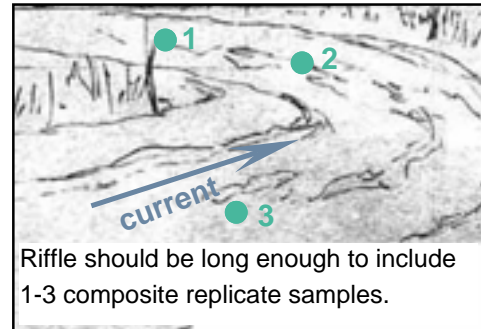
METHOD 1.A.2 NETS IN RIFFLES

COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET

On-Site Prep List

- 1) Fill in the top half of the *Collection Field Sheet*.
- 2) Measure a 200' segment that contains the riffle habitat and draw this segment on the back of the field sheet.
- 3) Identify your collection spots.
- 4) Wade to the downstream side of the first spot and get into position.

Replicate Sample Locations

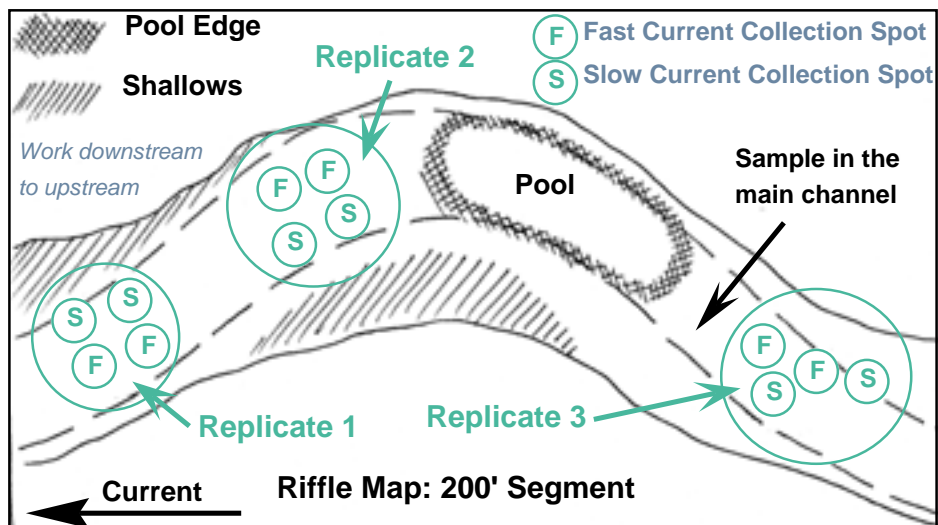


Tip:

Avoid shallow areas that might be dry at base (low) flow.

Tip:

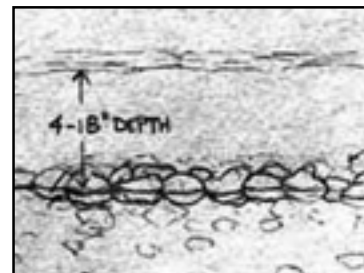
Avoid eddys behind rocks or your leg - they create an upstream flow that will allow your critters to escape.



A bit too large . . .



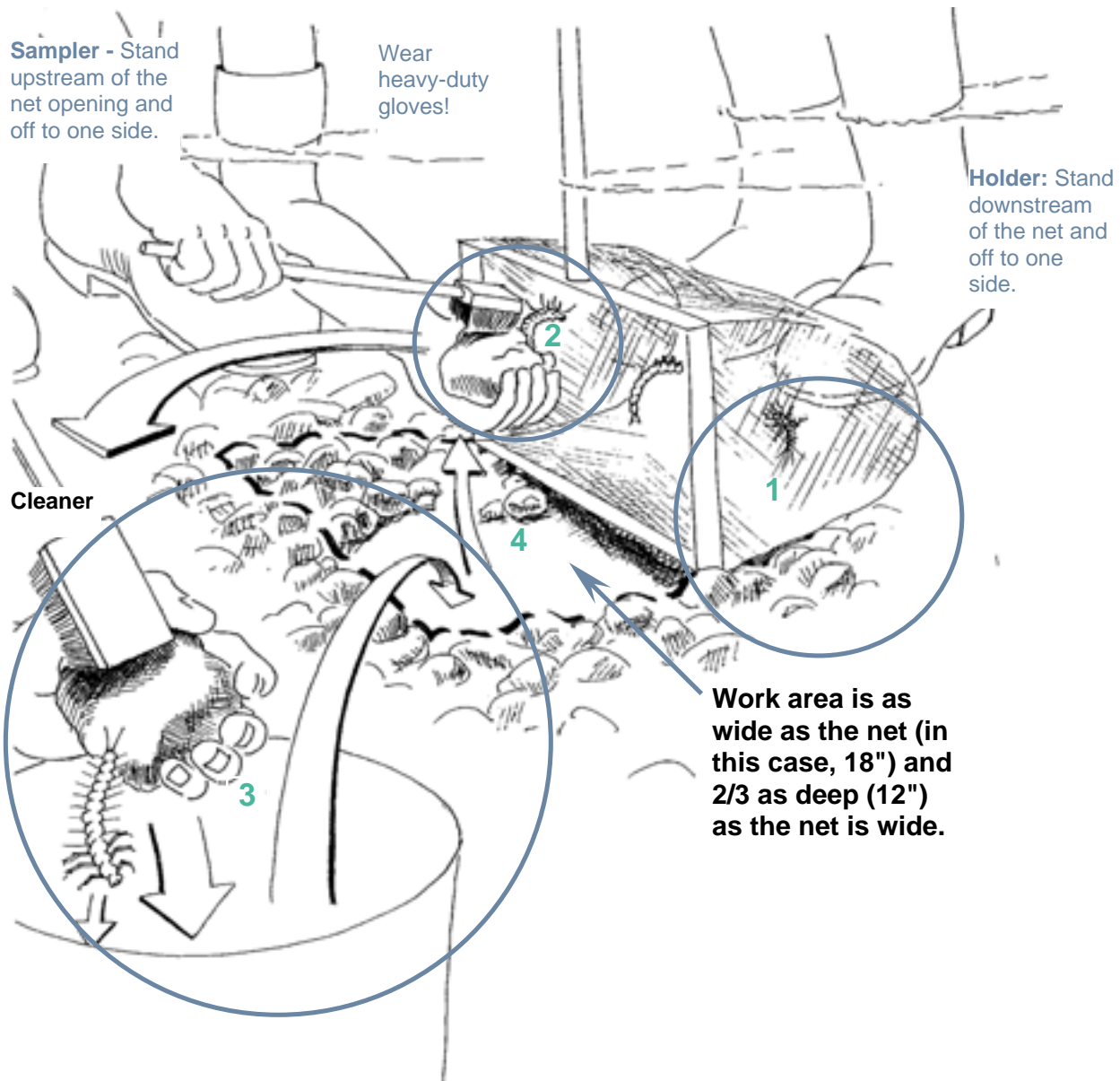
Ideal cobble =
2-10"



Sample cobble bottoms, no greater than 4 - 18" deep

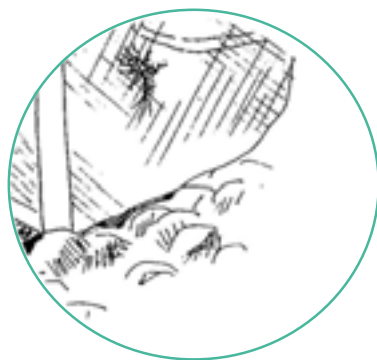
COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET

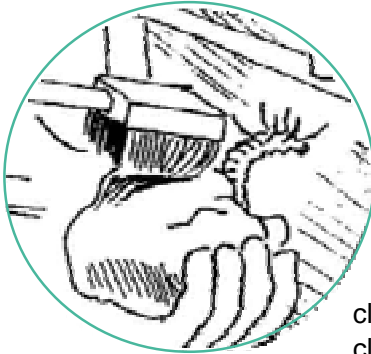
METHOD 1.A.2 NETS IN RIFFLES



Step 1: Place the Net

Holder: Place the net on the cobble bottom. Make sure it's seated so that there's no large gap under the frame. Be sure the current flows freely through the mesh. Avoid eddies.





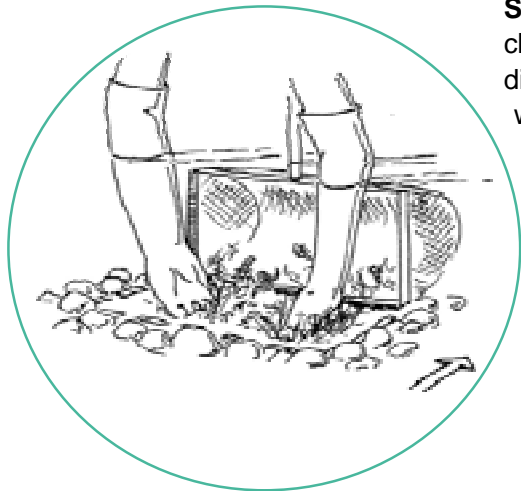
Step 2: Dislodge Critters from the Work Area

Sampler: Dislodge critters from all cobbles in the work area. Work under water. Pick up each cobbles and rub or brush with a soft-bristled brush toward the inside the net. Quickly inspect each for persistent clingers. If none, put the cobbles to one side, out of the work area. If there are some, keep brushing or pass it to the cleaner (if you've got one) for step 3 or place in a bucket for later cleaning.



Step 3: Get the Persistent Clingers

Cleaner: Working inside the bucket (1/3 full of water) brush, rub, (in one direction) or pick off the clingers from the cobbles. You may need tweezers.



Step 4: Get the Burrowers

Sampler: When all the cobbles in the work area are cleaned, gently dig into the bottom a few inches to dislodge burrowing critters. Too vigorous a technique will throw copious amounts of sand into the net, which you will regret later. Replace all cobbles in the work area.

Step 5: Lift the Net Out of the Water

It's not necessary to empty the net before collecting from the next spot unless it's plugged. Does it appear to be plugged up so that water cannot pass freely through it? Empty the net if you see that critters can crawl out, such as in relatively slow current.

Yes

No

Transfer enough sample from the net to the bucket to unplug the net and proceed to the next collection spot.



Proceed to the next collection spot to complete your composite replicate.

COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET: PRESERVING SAMPLES

METHOD 1.A.2 NETS IN RIFFLES

A ROAD MAP

Start here after you've collected each composite replicate (steps 1 - 5).

Step 6: Inspect the sample - set the vertebrates free!

Step 7: Empty the contents of the net into the bucket.

Step 8: Pick the critters off of the large debris.

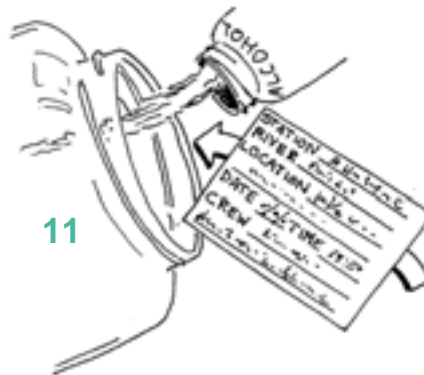
Step 9: Transfer the sample from the bucket to the sample container.

Step 10: Pick all remaining critters out of the net.

Step 11: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it. The label should contain the site #, replicate #, date, samplers initials, stream etc.



"We're free!"



Note:
See following pages for detailed steps.

Is the sample loaded with sand and gravel? If so, consider "swirling" it. See page 23 for instructions



A preserved sample

METHOD 1.A.2 NETS IN RIFFLES

COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET: HOW TO PRESERVE SAMPLES

DETAILED STEPS



After you've collected the first replicate, you have

- 1) a net with the sample in it, and
- 2) a bucket with cobble cleanings (and stuff taken out of the net to unplug it) and water.

The next steps depend on whether you are using a regular 5-gallon bucket or a sieve bucket. There are directions for both.

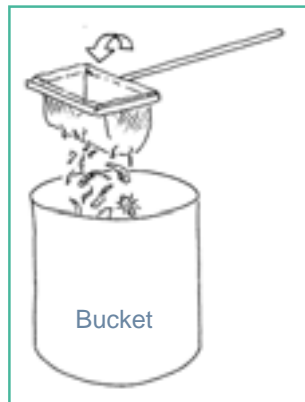
Is the sample loaded with sand and gravel? If so, consider "swirling" it. See page VII-23 for instructions



Step 6: Inspect the Sample

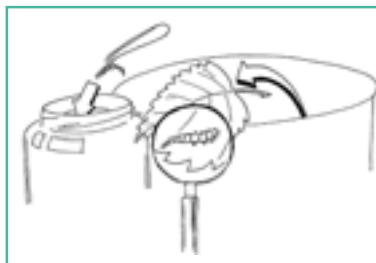
Is there a lot of sand and gravel in the net and the bucket? If it appears as though the sand and gravel will fill more than one sample container, you might consider re-sampling and doing it a little less vigorously. Or swirl the sample: see sidebar.

Set any vertebrates (small fish and amphibians) free!



Step 7: Empty the Contents of the Net Into the Bucket

Gather the contents into a clump in the net, grab the clump, and then turn the net inside out.



Step 8: Pick the Critters Off of the Large Debris

Pick out large debris (e.g. large leaves or sticks that won't fit into the the sample container) from the bucket. Pick all the critters off. Break open large rotten sticks and look for worms. Discard the large debris. Place critters into a labeled sample container.

DETAILED STEPS



Step 9: Transfer the Sample from the Bucket to the Sample Container

Strain the sample by pouring the contents of the bucket through a #30 sieve. Rinse out the bucket and repeat.

Scrape the sample from the sieve into the sample container. Hold a net under the sample container to catch any critters that don't make it into the jar. Pick any remaining critters out of the sieve and place them into the sample container.



Step 10: Pick All Remaining Critters Out of the Net

Place them into the sample container. A magnifying glass helps!



Step 11: Cover the Sample With Alcohol, Label, and Seal It

Samples will keep better if the sample container is filled with alcohol to within an inch of the top. If you haven't already, place a label - filled out in pencil or wax pencil (alcohol dissolves ink) in the jar. The label should contain the site #, replicate #, date, samplers initials, stream etc.

Repeat all steps 1 - 11 at each collection spot for each replicate.

METHOD 1.A.3 NETS IN RIFFLES

COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER



This method is the most rigorous method using a net in this manual. Because the Surber more precisely delineates an area of stream bottom (1 sq. ft.) it is considered more quantitative than the previous method (1.A.2) and will enable you to estimate the number of critters you collect per sample. This will be useful in looking at certain aspects of the benthic community described in Chapter VIII. It will also enable you to compare the number of organisms collected among your sample sites.

The following steps should be carried out by at least two people: one to hold the net (the “holder”) and the other to dislodge critters (the “sampler”). Another person and one to do a more thorough cleaning of the cobbles (the “cleaner”). The cleaner can also fill out the collection field sheet, and assist with sample processing and preservation.

Habitat Sampled	Riffle Bottom
Sampling Device	Surber Sampler
Level of Effort	Standardized by area
Quantitative?	Yes. Area delineated by the sampling device: 1.0 square feet sampled per replicate.
# Samples	3 single replicate samples

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- * Collection Field Sheets
- * 500-micron mesh Surber Sampler
- * Heavy-Duty Arm-length Gloves
- * 5-Gallon Buckets (2) and Sieve (#30 mesh)
- * Soft, Nylon Bristle Brushes
- * Forceps
- * Wide Mouth Sample Containers: sealable bottles or heavy-duty Zip-lockBags - 1 for each replicate
- * 90% de-natured Ethyl Alcohol
- * Labeling Tape and Pencils

2) Follow directions to the first site and find the riffle you will be sampling.

COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER

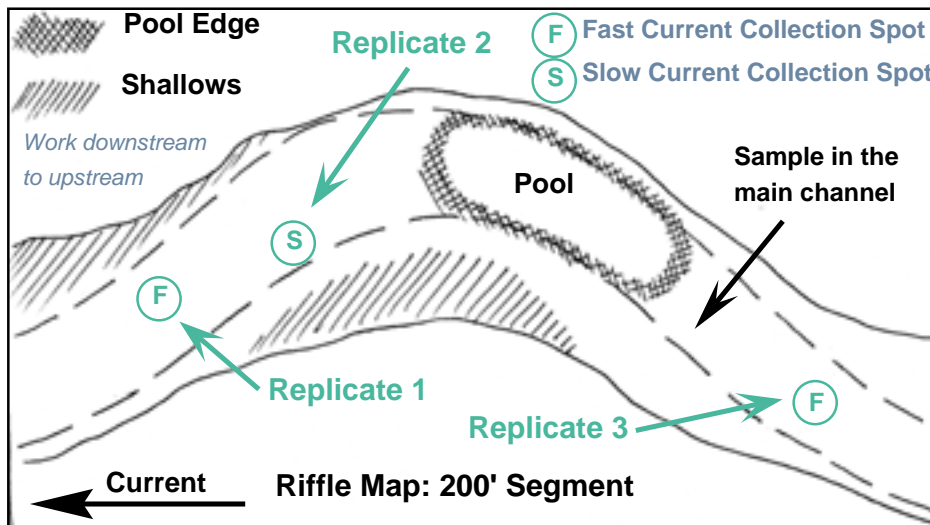
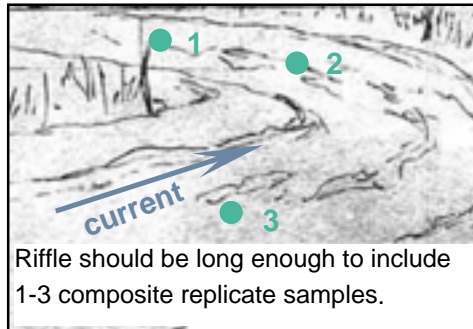
METHOD 1.A.3 NETS IN RIFFLES



On-Site Prep List

- 1) Fill in the top half of the *Collection Field Sheet*.
- 2) Measure a 200' segment that contains the riffle habitat and draw this segment on the back of the field sheet.
- 3) Identify your collection spots.
- 4) Wade to the downstream side of the first spot and get into position.

Replicate Sample Locations



Tip:
Avoid shallow areas that might be dry at base (low) flow.

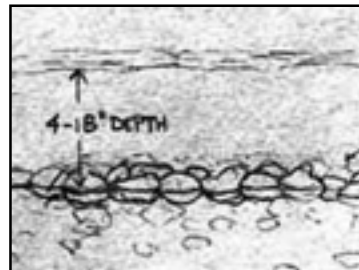
Tip:
Avoid eddys behind rocks or your leg - they create an upstream flow that will allow your critters to escape.



A bit too large . . .



Ideal cobble =
2-10"

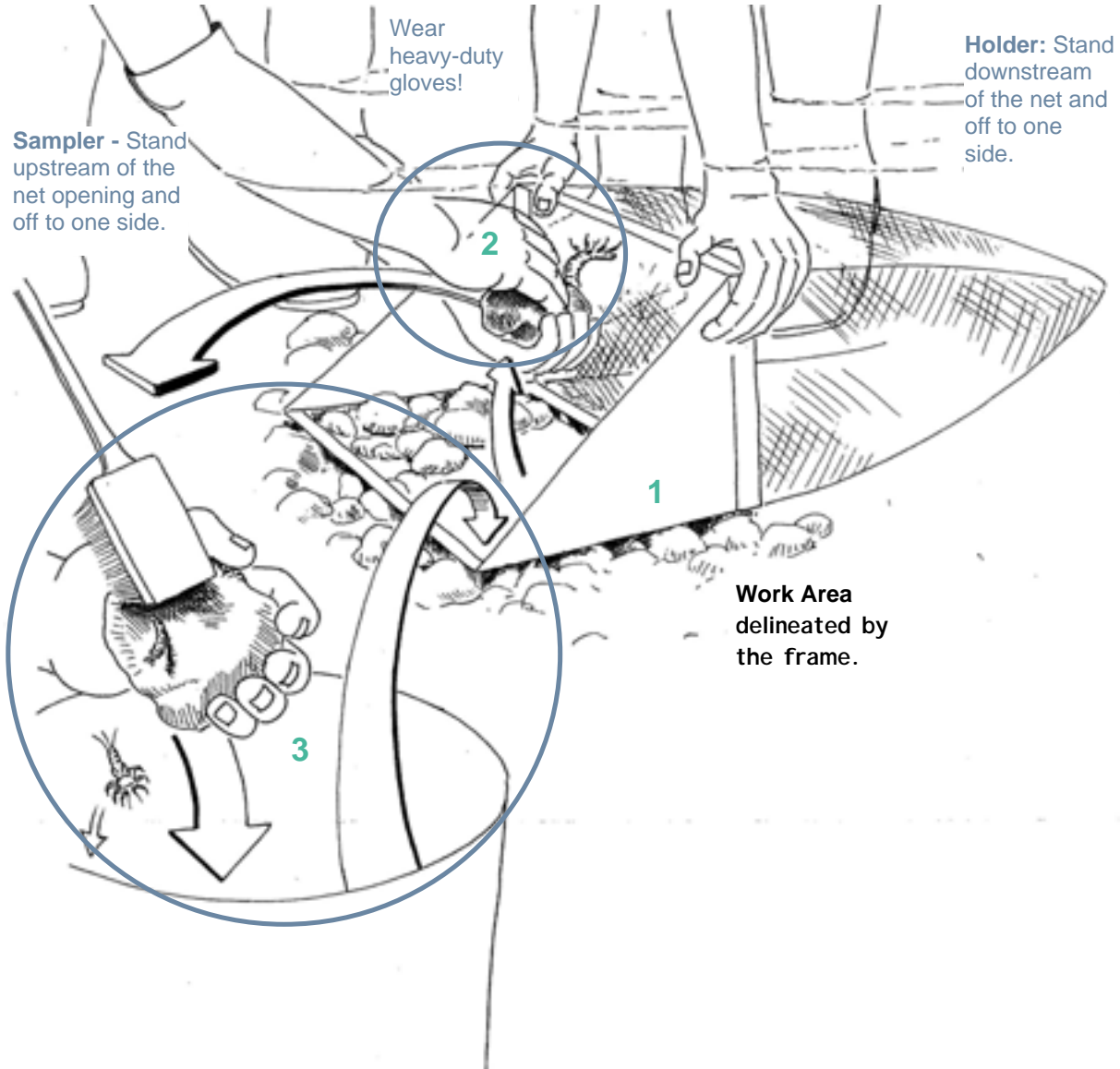


Sample cobble bottoms, no greater than 4 - 18" deep



METHOD 1.A.3
NETS IN RIFFLES

COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER



Step 1: Place the sampler on the bottom with the opening facing upstream

Holder: Brace the frame and hold it firmly on the stream bottom to prevent critters from getting under the net.



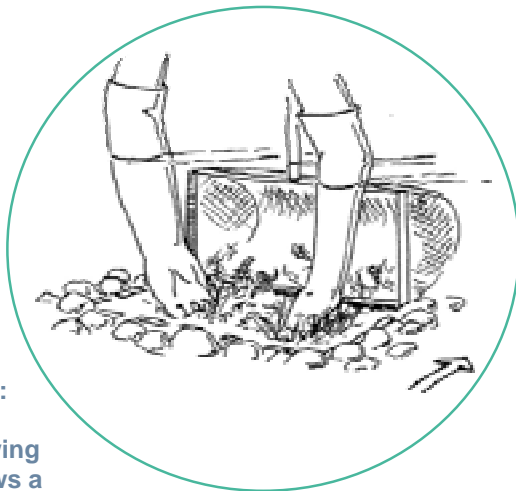
Step 2: Dislodge Critters from the Work Area

Sampler: Dislodge critters from all cobbles within the frame. Work under water. Pick up each cobble and rub or brush (with a soft-bristled brush) toward the inside of the net. Quickly inspect each for persistent clingers. If none, put the cobble to one side, out of the work area. If there are some, pass it to the cleaner (if you've got one) for step 3 or place in a bucket for later cleaning.



Step 3: Get the Persistent Clingers

Cleaner: Working inside the bucket (1/3 full of water) brush, rub, or pick off the clingers from the cobbles. You may need tweezers.



Note:
This drawing shows a metal frame net, but the step is the same for the Surber.

Step 4: Get the Burrowers

Sampler: When all the cobbles in the sampler frame are clean, gently dig (you can use a trowel or a rake) into the bottom several inches to dislodge burrowing critters. Too vigorous a technique will throw copious amounts of sand into the sampler, which you will regret later. Replace all cobbles in the work area.

Step 5: Lift the Net Out of the Water

Tilt the sampler up and out of the water while keeping the open end upstream. This helps to wash the organisms into the receptacle. Bring the sampler to the bank.

METHOD 1.A.3
NETS IN RIFFLES

COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER: How To PRESERVE SAMPLES

A ROAD MAP



Note:
See following pages for detailed steps.

Start here after you've collected each replicate (steps 1 - 5).

Step 6: Inspect the sample - set the vertebrates free!

Step 7: Empty the contents of the net into the bucket.

Step 8: Pick the critters off of the large debris

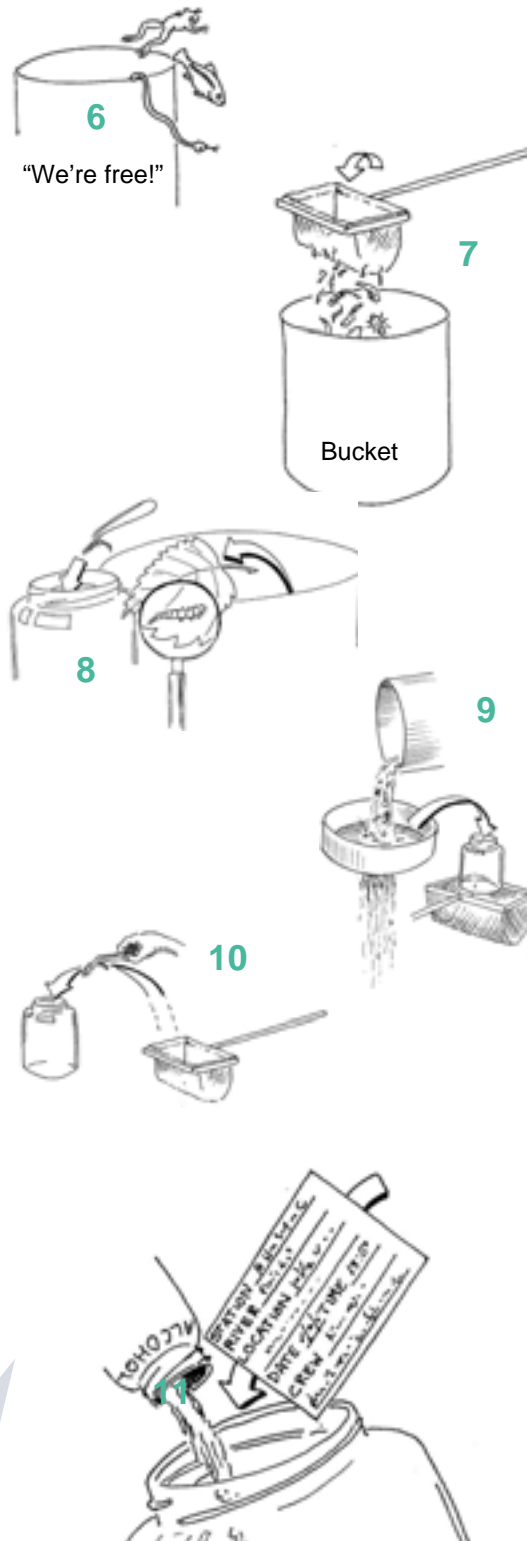
Step 9: Transfer the sample from the bucket to the sample container.

Step 10: Pick all remaining critters out of the net.

Step 11: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it.



A preserved sample



COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER: HOW TO PRESERVE SAMPLES

METHOD 1.A.3 NETS IN RIFFLES

DETAILED STEPS

After you've collected the first replicate, you have

- 1) a net with the sample in it, and
- 2) a bucket with cobble cleanings (and stuff taken out of the net to unplug it) and water.

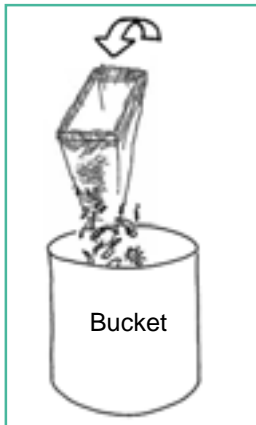
The next steps depend on whether you are using a regular 5-gallon bucket or a sieve bucket. There are directions for both.



Step 6: Inspect the Sample

Is there a lot of sand and gravel in the net and the bucket? If it appears as though the sand and gravel will fill more than one sample container, you might consider resampling and doing it a little less vigorously.

Set any vertebrates (small fish and amphibians) free!



Step 7: Empty the Contents of the Net Into the Bucket

On the bank, empty contents of Surber into a bucket or dishpan, by washing the sample to the narrow end of the net with the squirt bottle and water. Grab this end and turn it inside out into the bucket. Rinse the net into the bucket until all animals are removed.



Step 8: Pick the Critters Off of the Large Debris

Pick out large debris (e.g. large leaves or sticks that won't fit into the the sample container) from the bucket. Pick all the critters off. Discard the large debris. Place critters into the labeled sample container.

METHOD 1.A.3
NETS IN RIFFLES

COLLECTING A QUANTITATIVE SAMPLE WITH A SURBER SAMPLER

HOW TO PRESERVE SAMPLES

DETAILED STEPS



Step 9: Transfer the Sample from the Bucket to the Sample Container

Strain the sample by pouring the contents of the bucket through a #30 sieve. Rinse out the bucket and repeat.

Scrape the sample from the sieve into the sample container. Hold a net under the sample container to catch any critters that don't make it into the jar. Pick any remaining critters out of the sieve and place the



Step 10: Pick All Remaining Critters Out of the Net

Place them into the sample container. A magnifying glass helps!



Step 11: Cover the Sample With Alcohol, Label, and Seal It

Samples will keep better if the sample container is filled with alcohol to within an inch of the top. If you haven't already, place a label - filled out in pencil or wax pencil (alcohol dissolves ink) in the jar. The label should contain the site #, replicate #, date, samplers initials, stream etc.

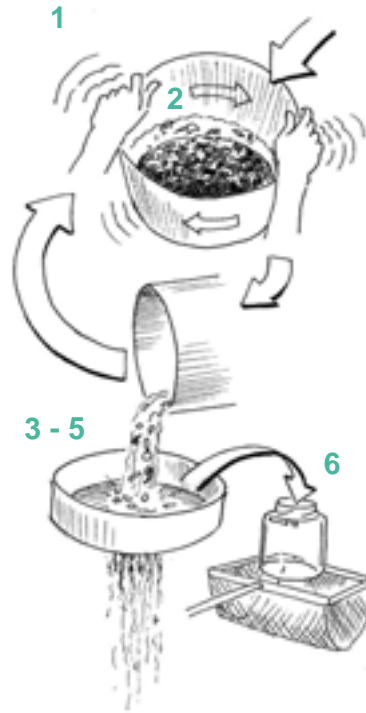
Repeat all steps 1 - 11 at each collection spot for each replicate.

HOW TO SWIRL SAMPLES



Swirling the Sample

- 1) Place the sample into a 5-gallon bucket about 1/3 full of water.
- 2) Use a swirling motion to create a whirlpool in the bucket. Organic debris and critters will get caught in the whirlpool.
- 3) Pour the water and organic debris into a #30 sieve, leaving the rocks, gravel and sand behind on the bottom of the bucket.
- 4) Replace the water and repeat the swirling 15-20 times until all that's left in the bucket is rocks, gravel and sand and the water you pour off has no debris in it.
- 5) Replace the water one more time and let the bucket sit for a few minutes. Carefully inspect the water in the bucket for floating or swimming organisms.
- 6) Scrape the trapped debris into the sample container.



Note: do this in the field only if you cannot fit the sample in the available containers.

B. Collecting Samples from Wadeable Low Gradient Habitats Using Nets

Low gradient, soft-bottom reaches provide very different habitat than riffles. Here the key is to find the structures that provide the most stable attachment surfaces. The most common are:

Vegetated bank margins

These consist of overhanging bank vegetation and submerged root mats attached to banks. The bank margins may also contain submerged, decomposing leaf packs trapped in root wads or lining the streambanks. This is generally a highly productive habitat in a muddy-bottom stream, and it is often the most abundant type of habitat. (EPA, 1997)

Snags and logs

These consist of submerged wood, primarily dead trees, logs, branches, roots, cypress knees and leaf packs lodged between rocks or logs. This is also a very productive muddy-bottom stream habitat. (EPA, 1997)

Aquatic vegetation beds

These consist of beds of submerged, green/leafy plants that are attached to the stream bottom. This habitat can be as productive as vegetated bank margins, and snags and logs. (EPA, 1997)

Silt/sand/gravel bottom

This includes sandy, silty, or muddy stream bottoms; rocks along the stream bottom; and/or wetted gravel bars. Algae-covered rocks (sometimes called Aufwuchs) may be present. This is the least productive of the four habitats, and it is always present in one form or another (e.g., silt, sand, mud, or gravel might predominate). (EPA, 1997)

COLLECTING A SEMI-QUANTITATIVE SAMPLE USING A NET

METHOD 1.B.1 NETS IN LOW GRADIENTS

Sampling each of these habitats involves a different procedure. The method we describe in this section is based on EPA's "muddy bottom" volunteer stream method (EPA, 1997). That method samples each habitat in proportion to its prevalence and combines the samples into a single composite. This guide departs from the EPA volunteer method in that we recommend that each habitat be sampled and preserved separately in order to preserve the information for each habitat. Results can always be combined during data analysis.



THE METHOD AT A GLANCE

Habitat Sampled	Banks, snags & logs aquatic vegetation, bottom
Sampling Device	Metal frame rectangular or D-net
Level of Effort	Standardized # of jobs
Quantitative?	Semi
# Samples	1 sample from each habitat

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- * Collection Field Sheets
- * Metal frame D-net (500-600 mm mesh)
- * Heavy-Duty Arm-length Gloves
- * 5-Gallon Bucket (2) and Sieve (#30 mesh)
- * Soft, Nylon Bristle Brushes
- * Plastic sugar scooper or ice-cream scooper
- * Forceps
- * Wide Mouth Sample Containers: sealable bottles or heavy-duty Zip-lock Bags - 1 for each replicate
- * 70-90% de-natured Ethyl Alcohol
- * Labeling Tape and Pencils

2) Follow directions to the first site and find the riffle you will be sampling.

METHOD 1.B.1
NETS IN LOW
GRADIENTS

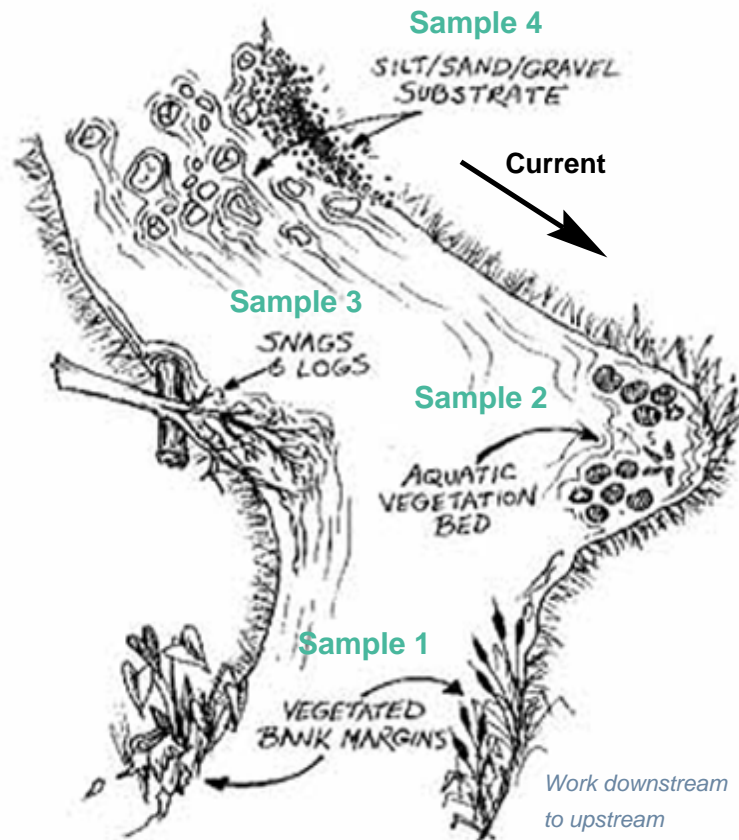
COLLECTING A SEMI-QUANTITATIVE SAMPLE USING A NET

On-Site Prep List

- 1) Fill in the top half of the *Collection Field Sheet*.
- 2) Measure a 200' segment that contains the habitats you will sample and draw this segment on the back of the field sheet.
- 3) Identify your collection spots, one for each habitat type present.
- 4) Wade to the downstream side of the first spot and get into position.

Tip:

Avoid shallow areas that might be dry at base (low) flow.



Source: EPA, 1997, Volunteer Stream Monitoring: A Methods Manual

COLLECTING A SEMI-QUANTITATIVE SAMPLE USING A NET

METHOD 1.B.1 NETS IN LOW GRADIENTS

About Jabs

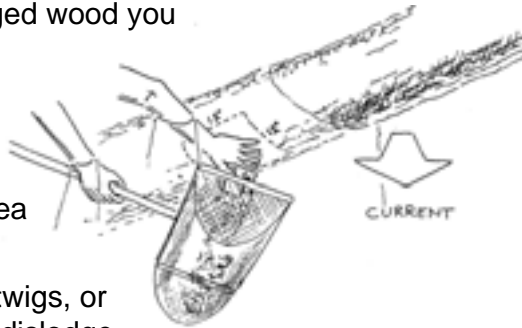
The basic unit of effort in this method is the “jab.” Each jab is about a foot in length and taken from each habitat type (described below) in proportion to its prevalence. (from EPA, 1997). Each sample is a specified number of jabs from a single habitat (see sidebar). In any case, be sure to record how many jabs you did in each habitat type.



Snags and Logs

Scrape 1 square foot of the snag or log per jab:

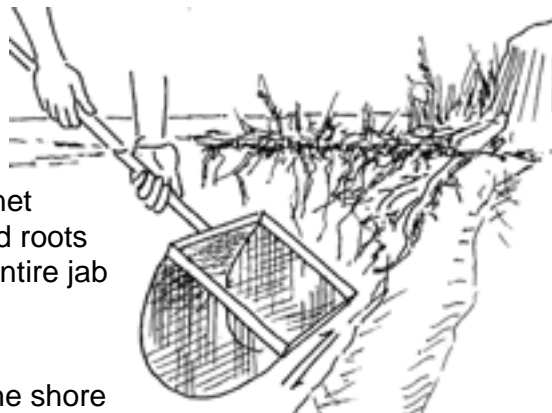
1. Hold the net with one hand under and downstream of the section of submerged wood you are sampling.
2. With the other hand (which should be gloved), rub or brush about 1 square foot of area on the snag or log.
3. Scoop organisms, bark, twigs, or other organic matter you dislodge into your net.
4. Bring the sample to the shore for processing and preserving.



Vegetated Banks

Wade to the downstream side of the bank and choose the specific place you will jab.

1. Jab the bank margin as many times as needed. Jab vigorously, with an upward 1-foot long motion, brushing the net against vegetation and roots along the bank. The entire jab motion should occur underwater.
2. Bring the sample to the shore for processing and preserving.



How Many Jabs?

- * **If all four habitats are plentiful**, jab the vegetated banks 10 times and divide the remaining 10 jabs among the remaining 3 habitats.
- * **If three habitats are plentiful** and one is absent, jab the silt/sand/gravel substrate (the least productive habitat) 5 times and divide the remaining 15 jabs among the other two.
- * **If only two habitats are plentiful**, jab the silt/sand/gravel substrate 5 times and the other 15 times.
- * **If some habitats are plentiful and others are sparse**, sample the sparse habitats to the extent possible, even if you can take only one or two jabs. Take the remaining jabs from the plentiful habitat(s).

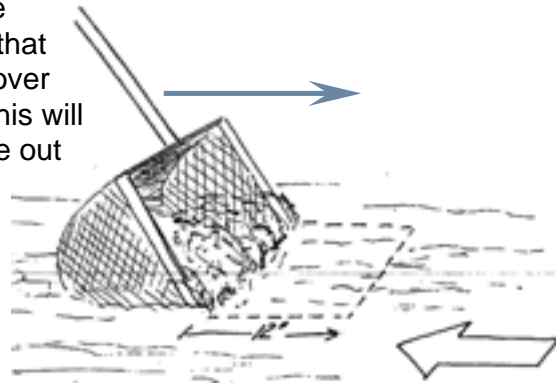
METHOD 1.B.1
NETS IN LOW
GRADIENTS

COLLECTING A SEMI-QUANTITATIVE SAMPLE USING A NET

Silt and Gravel Bottom

Wade to the downstream side of the area you will sample and choose the specific places you will sweep the net.

1. Place the net with one edge against the stream bottom and push it forward 1 foot (in an upstream direction) to dislodge the first few inches of silt, sand, gravel, or rocks.
2. To avoid gathering a netful of mud, periodically sweep the mesh bottom of the net back and forth in the water, making sure that water does not run over the top of the net. This will allow fine silt to rinse out of the net.
3. Repeat as many times as needed and bring the sample to the shore for processing and preserving.

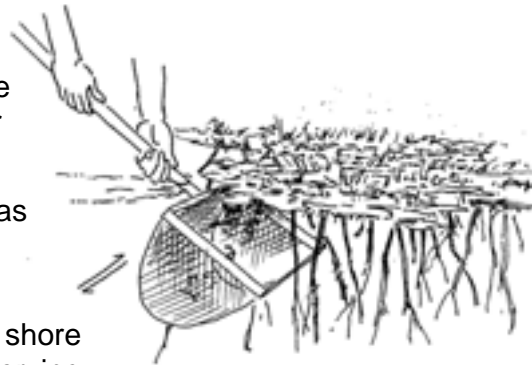


C

Aquatic Vegetation

Wade to the downstream of the section of the vegetation and choose the specific bed you will jab.

1. Jab the bank margin with the net: Jab vigorously, with an upward 1-foot long motion, brushing the net against or through the plant bed. The entire jab motion should occur underwater.
2. Repeat as many times as needed, leaving the sample in the net.
3. Bring the sample to the shore for processing and preserving.



COLLECTING A SEMI-QUANTITATIVE SAMPLE WITH A NET: PRESERVING SAMPLES

METHOD 1.B.1 NETS IN LOW GRADIENTS

A ROAD MAP

Start here after you've collected each replicate.

Step 1: Inspect the sample - set the vertebrates free!

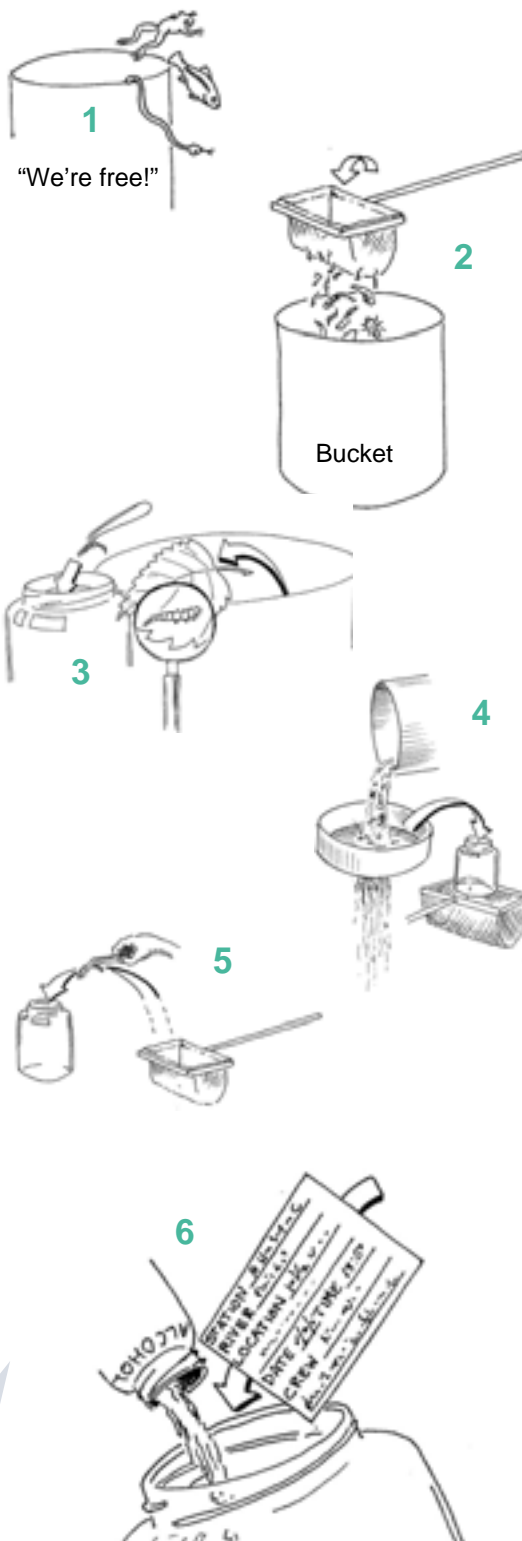
Step 2: Empty the contents of the net into the bucket.

Step 3: Pick the critters off of the large debris.

Step 4: Transfer the sample from the bucket to the sample container.

Step 5: Pick all remaining critters out of the net.

Step 6: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it. The label should contain the site #, replicate #, date, samplers initials, stream etc.



A preserved sample

C. COLLECTING SAMPLES WITH ARTIFICIAL SUBSTRATES

Artificial substrates are various types of devices which are placed on the river bottom or suspended in the water column that provide surfaces and spaces for the critters to colonize. They are left in place for anywhere from 4-8 weeks, retrieved, and the organisms collected from them. They can be used in a wide range of current velocities and depths.

Overview: What They Are and How They're Used

This guide covers two kinds of artificial substrates: multi-plate samplers and rock baskets. They can either be purchased or home-made.

Multi-plate Samplers

These consist of round or square hardboard or ceramic tiles stacked with spacers on an aluminum turnbuckle. The two most common types are the Hester-Dendy and the NY State Department of Environmental Conservation (NYDEC) samplers, shown in the margin.

Rock Baskets

The most common are cylindrical wire mesh baskets filled with 1.5 - 5.0 cm diameter rocks, similar to smaller cobbles found in the river (where you can collect them). Some people use golf balls because it's easier to estimate the surface area available for critters.

The type recommended and used by the U.S. EPA and other agencies for wadeable rivers is 11" X 6.5." A plastic coated cable with a loop on one end is threaded through the basket in order to anchor it to a steel rod or attach it to a cable running to a float.

We recommend a smaller and lighter 5" X 5" basket for deep rivers when you want to suspend it in the water column. We also recommend that rock baskets be coated with PVC to prevent rust.

How Artificial Substrates Are Used

The procedures in this chapter can be used with any type of artificial substrate. We suggest that you check with your state's aquatic biologist to see which type of artificial substrate is recommended and to get the specifications.

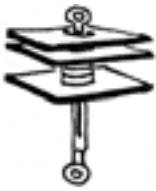
You'll encounter three types of river situations, described below. The placement procedures are different for each. However, in general, collecting benthic macroinvertebrates using artificial substrates involves:

1. placing them on the stream bottom (in wadeable waters) or hanging them in the water column (in deep waters)
2. retrieving them after a suitable colonization period, usually 5-6 weeks
3. removing the "tenants" from their short-term home.

Using artificial substrates requires teams of at least two people and, for non-wadeable sites, a boat.



Hester
Dendy



NY DEC



Rock Basket

Examples of Artificial Substrates

Rock Basket



Keys and Wallet



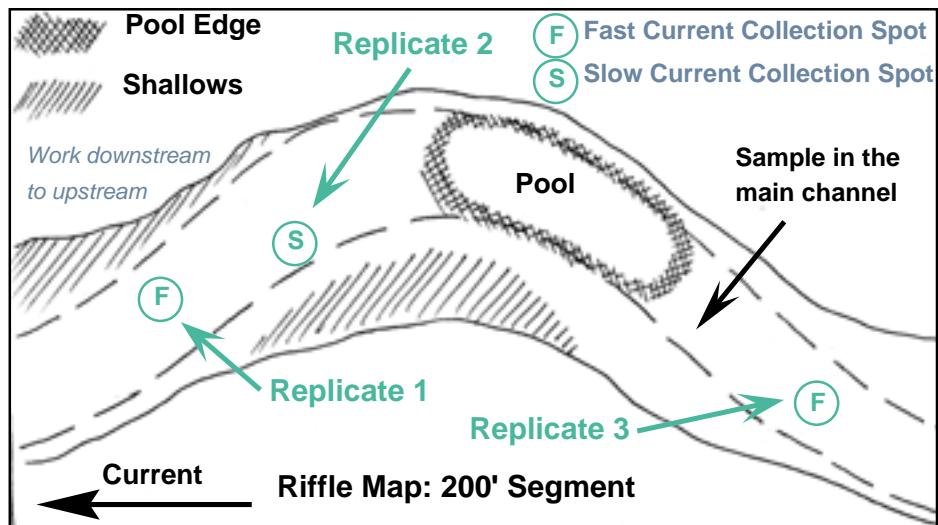
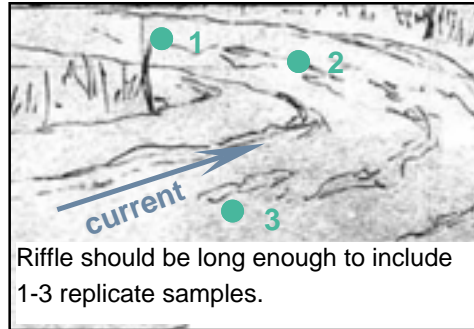
COLLECTING SAMPLES WITH ARTIFICIAL SUBSTRATES

PLACING THEM IN WADABLE WATERS

On-Site Prep List

- 1) Fill in the top half of the *Collection Field Sheet*.
- 2) Measure a 200' segment that contains the riffle habitat and draw this segment on the back of the field sheet.
- 3) Identify your placement spots.
- 4) Wade to the downstream side of the first spot and get into position.

Replicate Sample Locations



COLLECTING SAMPLES WITH ROCK BASKETS

METHOD 1.C.1 ROCK BASKETS IN RIFFLES

This method is considered quantitative because the level of effort is standardized as the surface area of the rocks in the baskets. It will enable you to enumerate the number of critters you collect per sample which will be useful in looking at certain aspects of the benthic community described in Chapter VIII. It will also enable you to compare the number of organisms collected among your sample sites.

THE METHOD AT A GLANCE

Habitat Sampled	Small cobbles, golf balls, fiberboard plates, or other materials placed on the stream bottom
Sampling Device	Rock Basket Artificial Substrate
Level of Effort	Standardized by area of basket
Quantitative?	Yes. Area delineated by the sampling device.
# Samples	3 single replicate samples

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- Collection Field Sheets
- Rock Baskets (3 per site)
- Rebar or comparable stake to anchor baskets and mark location
- Hammer
- Plastic-coated cable to attach basket to rebar
- Collection Net (500- 600 micron mesh)
- 5-Gallon Bucket and Sieve (#30 mesh)
- Soft, Nylon Bristle Brushes
- Forceps
- Wide Mouth Sample Containers: sealable bottles or heavy-duty Zip-lock Bags - 1 for each replicate
- 90% de-natured Ethyl Alcohol
- Labeling Tape and Pencils

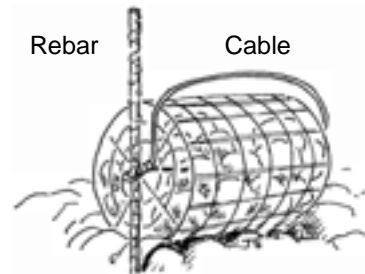
2) Follow directions to the first site and find the riffle you will be sampling.

COLLECTING SAMPLES WITH ROCK BASKETS

PLACING AND RETRIEVING IN WADABLE WATERS

Step 1:

Place sampler on the bottom. Hammer rebar into stream bottom and attach to basket with plastic-coated cable. Repeat at 2 other locations. Mark them with flagging tape so you can find them.

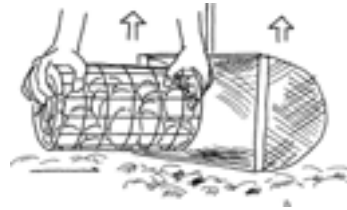


Four to Eight Weeks Later . . .

Approach the samplers from the downstream side.

Step 2:

Gently pull the basket off the bottom and use the collection net to catch organisms dislodged during this process.



Step 3:

Place the sieve in the bottom of the bucket and cover with water. Take apart the sampler in the bucket.

Remove the rocks from the basket and leave them in the sieve in bottom of the bucket. Place the basket in another bucket with water.



COLLECTING SAMPLES WITH ROCK BASKETS

PLACING AND RETRIEVING IN WADABLE WATERS

METHOD 1.C.1
ROCK BASKETS
IN RIFFLES

Step 4: Brush the rocks and the basket to get all the critters off. Pick any persistent clingers off with forceps.



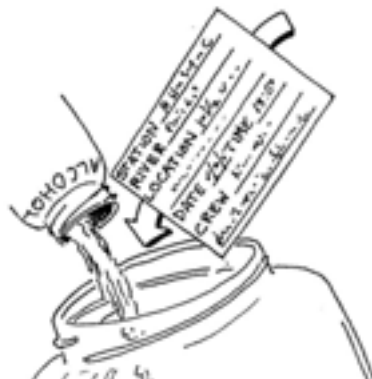
Step 5: Brush the rocks and the basket to get all the critters off.



Step 6: Transfer the sample from the bucket to the sample container. Pick any remaining critters out of the bucket and the net.



Step 7: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it. The label should contain the site #, replicate #, date, samplers initials, stream etc.



A preserved sample

COLLECTING SAMPLES WITH MULTI-PLATE SAMPLERS

This method is considered quantitative because the level of effort is standardized as the surface area of the plates that make up the sampler. It will enable you to estimate the number of critters you collect per sample which will be useful in looking at certain aspects of the benthic community described in Chapter VIII. It will also enable you to compare the number of organisms collected among your sample sites.

THE METHOD AT A GLANCE

Habitat Sampled	Multiple plates (fiberboard or ceramic) on eye-bolt placed on the stream bottom
Sampling Device	Multi-plate sampler (NY DEC or Hester-Dendy) artificial substrate
Level of Effort	Standardized by area of sampler
Quantitative?	Yes. Area delineated by the sampling device.
# Samples	3 single replicate samples

PRELIMINARIES

1) Assemble Collection and Processing Equipment and Supplies:

- * Collection Field Sheets
- * Multi-plate samplers (2) attached to concrete block (3 per site)
- * Collection Net (500- 600 micron mesh)
- * 5-Gallon Bucket and Sieve (#30 mesh)
- * Putty Knife
- * Forceps
- * Wide Mouth Sample Containers: sealable bottles or heavy-duty Zip-lockBags - 1 for each replicate
- * 90% de-natured Ethyl Alcohol
- * Labeling Tape and Pencils

2) Follow directions to the first site and find the riffle you will be sampling.



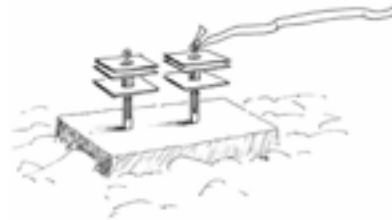
METHOD 1.C.2
MULTI-PLATES
IN RIFFLES

COLLECTING SAMPLES WITH MULTI-PLATE SAMPLERS

PLACING AND RETRIEVING IN WADABLE WATERS

Step 1:

Place sampler on the bottom.
Repeat at 2 other locations. Mark them with flagging tape so you can find them.

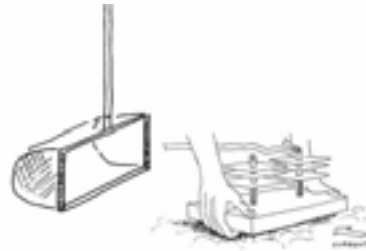


Four to Eight Weeks Later . . .

Approach the samplers from the downstream side.

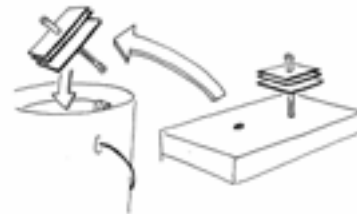
Step 2:

Gently pull the sampler off the bottom and use the collection net to catch organisms dislodged during this process.



Step 3:

Fill a 5-gallon bucket about 1/2 with water.
Remove the samplers from the block and place them in the bottom of the bucket..

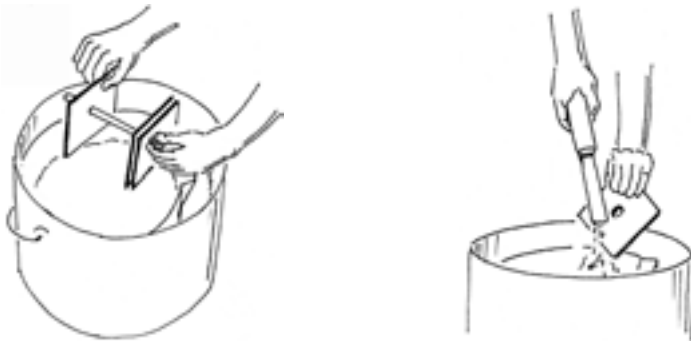


COLLECTING SAMPLES WITH MULTI-PLATE SAMPLERS

PLACING AND RETRIEVING IN WADABLE WATERS

METHOD 1.C.2
MULTI-PLATES
IN RIFFLES

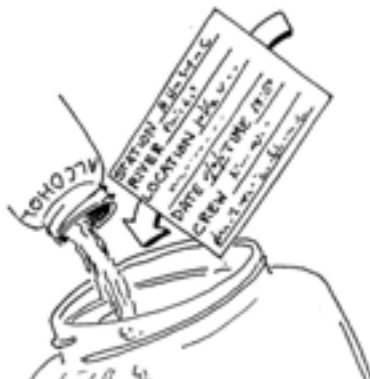
Step 4: Remove the plates from the eye bolt and scrape them with a putty knife. Scrape the block and the I-bolt too.



Step 5: Transfer the sample from the bucket to the sample container. Pick any remaining critters out of the bucket and the net.



Step 6: Cover the sample with alcohol, label w/ pencil, place the label in the container, and seal it.



METHOD 1.C.3
GENERAL
METHOD
ARTIFICIAL
SUBSTRATES
FOR
NON-WADABLE
WATERS

COLLECTING SAMPLES WITH ARTIFICIAL SUBSTRATES (MULTI-PLATE OR ROCK BASKET)

OPTIONS FOR PLACING THEM IN NON-WADABLE WATERS

Where the water is not wadeable, suspend the samplers (either a smaller rock basket or a multi-plate) in the water column using plastic-coated steel cable attached to a float or navigation buoy (bridge supports are sometimes used too) and a concrete block. Place samplers in the upper 0.3 - 1.0 meter of the water column, where light will reach the sampler. You will likely need a boat to place and retrieve the samplers.

Step 1: Attach the sampler to the float or buoy.

If the depth is greater than 2.0 meters, each of the samplers should be suspended between 0.3 and 1.0 meters below the water surface.

If the depth is between 0.5 and 2.0 meters, suspend each of the samplers midway between the water surface and the bottom.

Step 2: Lower the sampler and concrete block into the water.

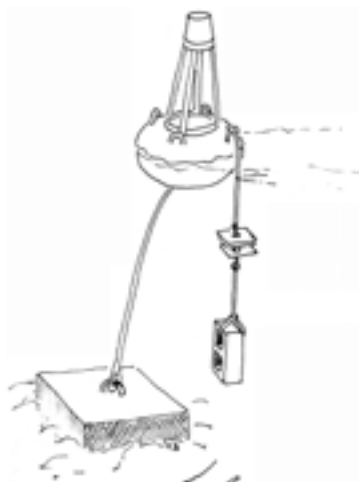
Step 3: Attach a plastic identification tag to the float or where the cable is attached to the buoy.

List your organization and a phone number. This will identify your organization in case someone finds your sampler before you do.



Float:

Sampler is hung between the float and a concrete block on the bottom.



Buoy:

Sampler and concrete block are hung from a buoy. The block keeps the sampler at the right depth (0.3-1.0 meters).

Note:

If you use a navigation buoy, you'll need permission from the agency that maintains the buoy.

If water levels will fluctuate by more than a few inches during the 5-week period of colonization, you should not use bridge supports to suspend your samplers because they will wind up too deep in the water column.

Be sure to place samplers so that cables are out of the navigation lanes!

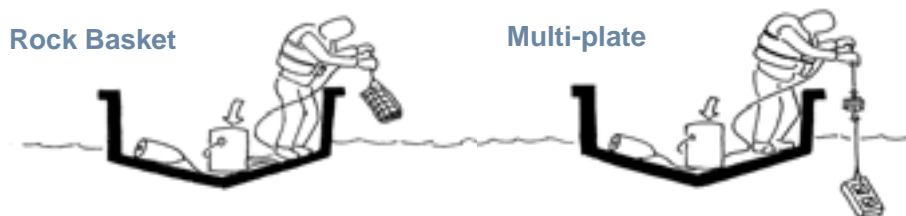
COLLECTING SAMPLES WITH ARTIFICIAL SUBSTRATES (MULTI-PLATE OR ROCK BASKET)

OPTIONS FOR PLACING THEM IN Non-WADABLE WATERS

Samplers should be retrieved after 4-6 weeks after placement. Consult with a biologist that knows your region to find out the optimal colonization period. You will need a boat and at least two people to retrieve the samplers from deep water.

Step 1: Remove the sampler from the water.

Detach the sampler from the buoy or float, slowly bring it to the surface and quickly place it in a 5-gallon bucket. Detach the cables from the sampler.

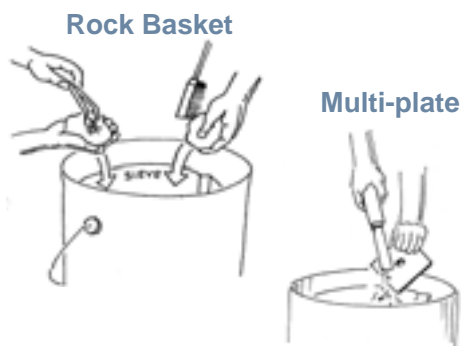


Step 2: Get the sample off the sampler.

Disassemble the sampler.

For Rock Baskets: Brush the rocks and the basket to get all the critters off.

For Multi-plates: Scrape the critters off the plate.



Step 3: Transfer the sample from the bucket to the sample container.

Pick any remaining critters out of the bucket and the net.



Step 4: Place a label, filled out in pencil or wax pencil (alcohol idssolves ink), in the jar. The label should contain the site #, replicate #, date, samplers initials, stream etc..



METHOD 1.C.3

GENERAL
METHOD
ARTIFICIAL
SUBSTRATES
FOR
NON-WADABLE
WATERS

COLLECTING SAMPLES: SAFETY!

Collecting benthic macroinvertebrate samples from moving water requires certain precautions and safety measures. We recommend the following:

- * Collect samples with a partner.
- * If you are in doubt as to your ability to safely collect a sample, don't do it! Be aware of your own physical limitations and the difficulty collecting samples at certain locations under certain conditions. Don't collect under difficult conditions.
- * Let someone know where you are going and when you expect to return.
- * Be careful when pulling off to the side of the road and leaving your car, so as not to endanger yourself or create a traffic hazard.
- * Consider leaving your wallet and keys in or around your car so you won't lose them in the river.
- * Bring a stick or pole along for balance when climbing down steep banks or wading.
- * Watch out for poison ivy – it likes stream banks!
- * High flows can turn even the most placid water into a raging torrent. Don't attempt to collect a sample if you feel the least bit of risk. Avoid dangerous situations. If there is an upstream dam that periodically releases water, the water level may rise swiftly. Be aware of the timing of such releases and avoid getting caught in the middle of the stream during a release. Inform the dam operators of your collection. If there is a conflict, ask if the release can be changed to accommodate you.
- * Collecting macroinvertebrate samples requires wading. Wear Waders! Bring a towel and a dry, warm change of clothes. If any of the people wading in the stream are not strong swimmers, the use of a personal floatation device is strongly recommended.
- * If sampling below a wastewater treatment plant, or in waters known to be polluted, wear rubber gloves and wash your hands after exposure.
- * If the stream bed is soft and mucky, test it with a pole or stick before putting your full weight on it.

QUALITY ASSURANCE

When Collecting Samples

The main quality assurance challenge is to make sure that the sample you collect is representative of the macroinvertebrate community living at the site. We recommend the following quality assurance measures for collection:

- 1) **Be Careful When Selecting Specific Collection Spots:** Be sure that you select good collection spots in the riffle. Make sure that they are predominantly cobble and have the required current velocity.
- 2) **For Methods 1.A.2 and 1.A.3 Collect 3 Replicates:** This helps assure the representativeness of the samples collected and is built into the procedure. If this is not practical, collect as many as you can.
- 3) **For Method 1.A.2 Be Sure To Collect Composite Samples:** Each sample should be a composite of 4 collections at each site. This helps assure the representativeness of the samples collected. This is built into the procedure.
- 4) **For Method 1.A.1, Collect Three Replicate Samples:** Three replicate samples collected at each site assure that the organisms you collect are representative of those living at the site. This is built into the Intensive Collection procedure.
- 5) **Collect Duplicate Samples:** Another team or, better yet, a professional aquatic biologist should collect samples at 10% of the collection sites, at the same time (within a day or so) as the volunteer team.
- 6) **Have As Few Collecting Teams As Practical:** The more people that collect the samples, the greater the chance for errors and inconsistent level of sampling effort.