INVERTEBRATE COMMUNITY SAMPLING PROTOCOL FOR STREAM MONITORING SITES

I. PURPOSE

To describe methods used in the collection of stream invertebrates for the purpose of developing biological criteria used in assessing water quality.

II. REFERENCES

A. Source Documents

U.S. Environmental Protection Agency (USEPA). 1994. Environmental Monitoring and Assessment Program - Surface Waters and Region 3 Regional Environmental Monitoring and Assessment Program: 1994 pilot field operations and methods manual for streams. U.S. Environmental Protection Agency, Environmental Monitoring Systems Laboratory. Cincinnati, OH. EPA/620/5-94/004.

Barbour, M. T., J. Gerritsen, and J. S. White. 1996. Development of the Stream Condition Index (SCI) for Florida. Florida Department of Environmental Protection, Tallahassee, Florida. 105 pp.

B. Other References

U.S. Environmental Protection Agency (USEPA). 1996. Biological Criteria: Technical Guidance for Streams and Small Rivers. Revised Edition. Office of Water, Washington DC. EPA/822/B-96/001.

U.S. Environmental Protection Agency (USEPA). 1997. Revision to Rapid Bioassessment Protocols for Use in Streams and Rivers (Draft). Office of Water, Washington D.C. EPA/841/D-97/002.

III. SCOPE/LIMITATIONS

This procedure applies to all site visits in which stream invertebrates are to be collected for the development of biological criteria and/or the assessment of water quality.

IV. DEFINITIONS

Integrated monitoring: A stream monitoring technique to assess water quality using chemical, biological and physical indicators.

Environmental Monitoring and Assessment Program (EMAP): U.S. Environmental Protection Agency program designed to determine the status, extent, changes, and trends in the condition of our national ecological resources on regional and national scales.

Biological Criteria: Narrative expressions or numerical values that describe the reference biological integrity of a specified habitat. Biological criteria are the benchmarks for judging the condition of aquatic communities.

Qualitative Multihabitat Sample (QMH): A method of sampling invertebrates which involves sampling a variety of invertebrate habitats, including the following: rocky substrates, including riffles and runs, submerged and emergent aquatic vegetation, undercut banks, overhanging vegetation, woody debris, and leaf packs.

Intensive Watershed Monitoring: A watershed monitoring plan designed to assess the aquatic health of major watersheds through intensive biological and water chemistry sampling. This intensive approach allows assessment of watersheds for aquatic life, aquatic recreation, and aquatic consumption use support of the state's streams in each of the state's 84 major watersheds on a rotating 10 year cycle.

V. GENERAL INFORMATION

The methods described herein are to be applied to all wadeable and non-wadeable streams included in the MPCA's integrated stream condition monitoring program. This document is intended to be used in concert with the documents indicated in the box below if any of the described situations apply. For most efficient use of time and resources, crew leaders should be able to easily communicate with each other and with crews sampling for fish to ensure safe access to sampling locations, and to avoid duplication of effort.

Data generated from samples collected using the described method can be used for any of the following reasons: 1) assessment of aquatic life uses as part of intensive watershed monitoring program, 2) data supplementation to aid the stressor identification process, 3) development of regional biological criteria, 4) calibration of biological criteria.

NOTE

SOP1 - Site Reconnaissance: A site reconnaissance should be done in the fall or spring prior to sampling.

SOP2 - Chemical Assessment: A chemical assessment, including the collection of field and laboratory chemistry samples, should be done by the fish crews. Macroinvertebrate field crews are responsible for collecting field chemistry unless a site is only being sampled for macroinvertebrates, in which case a laboratory sample should also be collected.

SOP3 – Minnesota Stream Habitat Assessment: A habitat assessment should be conducted by the fish crews unless a site only being sampled for macroinvertebrates, in which case the macroinvertebrate field crew should conduct the assessment. If a habitat assessment is to be done during the same visit as an invertebrate collection, the invertebrate collection should be done first.

VI. REQUIREMENTS

A. Qualifications of Crew Leaders

A crew leader must be a professional aquatic biologist with a minimum of a Bachelor of Science degree in biology with an aquatic entomology, invertebrate zoology, fisheries, or closely related specialization, or equivalent experience in a related field. Additionally, they should have previous professional experience working as a field biologist, including sampling macroinvertebrates, and conducting habitat assessments. Field crew leaders must possess excellent map reading skills, have a demonstrated proficiency in the use of a GPS (Global Positioning System), and have good interpersonal skills for dealing with landowners, and other interested stakeholders..

B. Qualifications of field technicians/interns

A field technician/intern must have at least one year of college education and had coursework in environmental and/or biological science.

C. General Qualifications

All personnel conducting this procedure must have excellent map reading skills and a demonstrated proficiency in the use of a GPS receiver and an orienteering compass. Because sites may be located miles from the nearest vehicle assessable road, it is often necessary to wade through streams and/or wetlands, canoe, or hike for long distances to reach a site. Personnel conducting this procedure must have the physical ability to accomplish this.

II. RESPONSIBILITIES

A. Field Crew Leader

Ensures that data generated using this procedure meet the standards and objectives of the integrated condition monitoring program. Carries out the procedures outlined in the action steps.

B. Technical personnel

Carries out the procedures outlined in the action steps, including maintenance and stocking of equipment, date collection and recording.

VII. QUALITY ASSURANCE AND QUALITY CONTROL

Compliance with this procedure will be maintained through annual internal reviews. Technical personnel will conduct periodic self-checks by comparing their results with other trained personnel. Calibration and maintenance of equipment will be conducted according to the guidelines specified in the manufacturer manuals.

In addition to adhering to the specific requirements of this sampling protocol and any supplementary site specific procedures, the QA/QC requirements for this protocol are as follows:

A. Control of Deviations
Deviations from the procedure shall be sufficiently documented to allow repetition of the activity as actually performed.

B. QC Samples
Ten percent of all sites sampled on any given year are resampled as a means of determining sampling error.

C. Verification
The field crew leader will conduct periodic reviews of field personnel to ensure that technical personnel are following the procedures according to this SOP.

IX. TRAINING

A. All personnel, including experienced staff, will receive annual instruction from a trainer designated by the program manager.

B. Training activities will include instruction in the field as well as a field test to ensure that personnel can implement this procedure.

C. The field crew leader will provide instruction in the field to untrained personnel, such as interns and technicians, to ensure they can effectively execute this procedure.

X. ACTION STEPS

A. Equipment List

Ensure that all of the following items are presents before implementing this procedure:
Two D-frame dipnets with 500 micron mesh nets, equivalent to Wildco, turtox design
Two sieve buckets with 500 micron sieves
Stream Invertebrate Visit FormElectronic or hardcopy of information included in Stream Verification Form, along with aerial imagery and copies of 1:24,000 USGS topographical map highlighting stream reach.
Minnesota Atlas and Gazateer (Delorme)
Pencils
Permanent/Alcohol proof marker
Labeling tape
Internal and External invertebrate sample identification labels
100% reagent alcohol, adequate volume to preserve four days worth of samples, ca. 10-15 gallons
Waterproof notebook
Chest-high waders
Rain-gear
Jars or bottles in which sample is to be preserved; preferably non-breakable synthetic, minimum 1 litre capacity
Box or crate to store sample bottles
Canoe or Kayak if needed
Backpack

B. Method

The multi-habitat method entails collecting a composite sample from up to five different habitat types. The goal of this method is to get a sample representative of the invertebrate community of a particular sampling reach, it is also to collect and process that sample in a time and cost effective manner. For that reason the habitats described below are relatively non-specific, being chosen to represent broad categories rather than microhabitats. Every broad category includes numerous microhabitats, some of which will not be sampled. It is to the discretion of the sampler which microhabitats are most representative of a reach. As a general rule, sample in manner that reflects the most common microhabitat of any given broad habitat category. The habitats to be sampled include:

Hard bottom (riffle/cobble/boulder)
This category is intended to cover all hard, rocky substrates, not just riffles. Runs and wadeable pools often have suitable “hard” substrates, and should not be excluded from sampling. The surfaces of large boulders and areas of flat, exposed bedrock are generally quite unproductive, avoid including these habitats in the sampling area if possible. This is a general rule, if a particular stream has productive exposed bedrock, or boulder surfaces, those habitats should be considered sampleable.

Aquatic Macrophytes (submerged/emergent vegetation)
Any vegetation found at or below the water surface should be considered in this category. Emergent vegetation is included because all emergent plants have stems that extend below the water surface, serving as suitable substrate for macroinvertebrates. Do not sample the emergent portion of any plant.

Undercut Banks (undercut banks/overhanging veg)
This category is meant to cover in-bank or near-bank habitats, shaded areas away from the main channel that typically are buffered from high water velocities.

Snags (snags/rootwads)
Snags include any piece of large woody debris found in the stream channel. Logs, tree trunks, entire trees, tree branches, large pieces of bark, and dense accumulations of twigs should all be considered snags. Rootwads are masses of roots extending from the stream bank.

Leaf Packs
Leaf packs are dense accumulations of leaves typically present in the early spring and late fall They are found in deposition zones, generally near stream banks, around logjams, or in current breaks behind large boulders.

B. Method (continued)

Sampling consists of dividing 20 sampling efforts equally among the dominant, productive habitats present in the reach. If 2 habitats are present, each habitat should receive 10 sampling efforts. If 3 habitats are present, each habitat should receive 7 sampling efforts. If a productive habitat is present in a reach but not in great enough abundance to receive an equal proportion of sampling efforts, it should be thoroughly sampled and the remaining samples should be divided among the remaining habitat types present.

A sample effort is defined as taking a single dip or sweep in a common habitat. A sweep is taken by placing the D-net on the substrate and disturbing the area directly in front of the net opening equal to the net width, ca. 1ft². The net should be swept several times over the same area to ensure that an adequate sample is collected. Each effort should cover approximately .09m² of substrate. Total area sampled is ca. 1.8m².

Once a site reach has been found or newly established, invertebrate sampling should follow. If a habitat assessment is to be conducted and/or a chemistry sample collected it should follow invertebrate sampling.

NOTE
Before leaving the vehicle be sure that the following equipment is brought to the site: two d-frame dipnets, two sieve buckets, site visit form, site file, GPS receiver, camera, sample bottles, multi-meter, and chemistry sample bottles if needed.

1**. Assessing Stream Habitats** - Before sampling can begin, the Crew Leader and field tech must determine which habitats are present in the reach. This should be a cooperative effort. This is done by walking the length of the stream and determining which productive habitats dominate the stream reach. A site visit form should be filled out during this process or immediately following sample collection. Ideally the stream should be viewed from the top of the stream bank, but this is generally the exception rather than the rule. For this reason, great care must be taken to walk gingerly along the stream edge, or any streamside exposed areas. If this is not possible, stay to one side of the stream so as to disturb as little substrate as possible.

B. Method (continued)

NOTE
Since sampling should be conducted in a downstream to upstream fashion, it will save time to start the initial visual inspection of the stream from the upstream end of the sampling reach, and walk downstream. This will allow you to start sampling at the down stream end of the reach as soon the inspection is completed.

It can be difficult to estimate total stream coverage of certain habitats due to their appearance as linear or two dimensional features. Undercut banks and overhanging vegetation can appear as linear features despite their depth, while snags, woody debris, vegetation mats, and emergent vegetation can appear flat despite their three dimensional nature. For these reasons best professional judgment must be used to determine what level of effort is adequate to equal one "sample effort" for any given substrate. Keep in mind that this method is considered qualitative, rulers and grids are not necessary to effectively implement this procedure. Following are some suggestions as to how approach each habitat for the perspective of

Hard bottom: Riffles and rocky runs are basically two dimensional areas, and should be thought of as such when trying to determine how dominant the riffle habitat is in a stream. It must be kept in mind that riffles are often the most productive and diverse habitat in the reach, relatively speaking. The field personnel must be careful to not oversample riffles. The purpose of this method is to get a representative sample. Sampling in this habitat type is relatively simple. The D-net should be placed firmly and squarely on the substrate downstream of the area to be sampled. If the water is shallow enough, the area directly in front of the net should be disturbed with the hands, taking care to wash large rocks off directly into the net. If the water is too deep for this, kicking the substrate in front of the net is adequate. Watch for stoneflies and mayflies trying to crawl out of the net.

Vegetation: Aquatic vegetation is either completely submerged, mostly submerged and partially floating on the waters surface, or partially submerged and mostly extended above the waters surface. Things like pondweed, coontail, and milfoil tend to clump and float at the waters surface. These types of plants should be sampled with an upward sweep of the net. If the net fills with weeds, the weeds should be hand washed vigorously or jostled in the net for a few moments and then discarded. Emergent plants such as reed canary grass and various plants in the rush family, should be sampled with horizontal and vertical sweeps of the net until it is felt that the area being swept has been adequately sampled. Plants like floating bur reed, and water celery tend to float in long strands with the current. They can be floating on the surface of completely submerged. These plants should be sample as emergent plants with horizontal and vertical sweeps in a downstream to upstream motion.

Undercut banks/ Overhanging Vegetation: Undercut banks and overhanging vegetation follow the line of the stream bank. Undercut banks can vary in how undercut they are. An additional problem is that many banks appear undercut, but when investigated prove not to be. For these reasons banks should be prodded to determine how deeply they are undercut. Overhanging vegetation should be treated the same way. Sampling should consist of upward thrusts of the net, beating the undercut portion of the bank or the overhanging vegetation, so as to dislodge any clinging organisms.

Snags: Snags and rootwads can be large or small, long or wide, simple or twisted masses of logs or twigs that don't have any consistent shape. Best professional judgment must be used to determine what a "sampling effort" is. Approximating the amount of sampleable surface area is a sensible method with larger tree trunks or branches. Masses of smaller branches and twigs must be given a best guess. Given their variable nature, there is not one best method for sampling snags. Using something like a toilet brush or kitchen brush works well for large pieces of wood, whereas kicking and beating with the net works best for masses of smaller branches. The person taking the sample must determine the best method for each particular situation.

Leaf packs: Leaf packs are simple, but messy to sample. One square foot of leaf pack surface area that has two cubic feet of leaf underneath should be sampled near the surface. Whereas a shallow leaf pack can be sampled in it's entirety. Sweeping to the bottom of every leaf pack could create a disproportionately large amount of sample volume being collected for relatively small sample area. In most situations leaf packs will not be dominate enough to be included in a sample. If leaf packs are sampled, it is suggested that time be spent streamside washing invertebrates off of leaves and discarding the leaves, as a leaf pack sample can easily become overwhelmingly large.

B. Method (continued)

2. **Sampling the macroinvertebrates** - After the number of productive sampleable habitats have been determined, the sampling team should proceed in a downstream to upstream manner, sampling the various habitats present.

NOTE

In order to get complete samples, the contents of the D-net should be emptied into a sieve bucket frequently. This prevents the back flow of water resulting from a clogged net. In larger streams it is convenient for each sampler to have a sieve bucket. This allows samplers to sample independent of each other, avoiding frequent stream crossings which can alter the stream bed.

NOTE

While sampling it may become necessary to clean the sample of muddy, fine sediment. This can be done by filling the sieve bucket with clean water and allowing the resulting mucky water to drain. Care must be taken not twist and turn the bucket to much, this creates a washing machine action which separates insects from their delicate parts quite effectively.

3. **Preserving the sample** - Once sampling is complete the sample material should be preserved as quickly as possible. Transfer the sample material from the sieve bucket to the sample containers. Sample containers should contain no more than 30% of their volume as wet weight. Fill sample containers with 100% reagent alcohol to a level that ensures a final alcohol concentration of at least 70%. Be sure to thoroughly clean the bucket and sampling nets of all invertebrates. The use of forceps might be necessary to dislodge some of the smaller organisms.

4. **Labeling the sample** – Fill out internal and external sample labels for each sample container using preprinted sample labels (see attachement). Be sure to use water and alcohol proof writing medium.

XI. REQUIRED RECORDS

Stream Invertebrate Visit Form

A. The Stream Invertebrate Visit Form should be filled out during the streamside survey, or notes should be taken on field note books and transferred to visit forms. This information will be placed in the biological database.

MPCA Stream Monitoring Program

STREAM INVERTEBRATE VISIT FORM

Stream Name:		**Date:**
Field Number:	**County:**	**Crew:**
Water Chemistry	Tape Down: ___._____ (1/100ths ft)	Location: _____

Time: (24 hr) ___:___ Air Temp:_____ (°C) Water Temp:_____ (°C) Conductivity: _____ (umhos@25°C)

DO: _____(mg/L) DO % Saturation:_____ pH:_____ Secchi -Tube: _____ (cm)

Water Level: Normal Below _____ (m) Above _____ (m) Color _____ (pcu)

*****If Flagging is not found or if establishing a new site, fill out GPS info*****

Coordinates	**LATITUDE**	**LONGITUTDE**	**Time:**
Field GPS:	____._____	____._____	**Name:**

Notes:

Stream Classification Information

Flow	Flow over riffle(s)	High / Med / Low / NA	Channel	Excavated, trapezoidal channel	%
	Flow at reach constriction	High / Med / Low / NA		Shallow excavation, channelized wetland	%
	Flow over run	High / Med / Low / NA		Natural channel	%
	General flow pattern	High / Med / Low / NA	Vegetation	Emergent, aquatic vegetation in channel	Ext / Mod / Sparse / NA
	Intermittent sections	Yes / No		Emergent, aquatic vegetation along bank	Ext / Mod / Sparse / NA
Habitat	Riffle (with flow) present in reach c			Floating or submerged aquatic vegetation	Ext / Mod / Sparse / NA
	Riffle (with flow) present outside of reach c (riffles do not include riprap associated with bridges or bank stabilization)			Loosely attached filamentous algae	Ext / Mod / Sparse / NA
				Firmly attached algae or submerged veg	Ext / Mod / Sparse / NA
	Dominant invertebrate habitat (circle two) Riffle | Rocky Run-Pool | Aquatic Macrophyte | Bank-Overhanging Veg | Wood | Leaf				
Substrate	Dominant Run Substrate	bedrock / boulder / cobble / gravel / sand / silt			
	Dominant Pool Substrate	bedrock / boulder / cobble / gravel / sand / silt			
	Dominant Substrate receiving flow	bedrock / boulder / cobble / gravel / sand / silt			
	Dominant Substrate in reach	bedrock / boulder / cobble / gravel / sand / silt			

c	Stream displays a typical riffle-run pool morphology c adequate flow to maintain riffle organisms c inadequate flow to maintain riffle organisms
c	Stream has adquate flow to maintain riffle organism, but does not have suitable coarse substrate to support these assemblages (riffles, rock substrate in runs or pools)
c	Stream has adquate flow to maintain riffle dwelling organism, woody debris has replaced rocks as primary coarse substrate
c	Stream is low gradient, stream bed is predominately fine substrate, inadequate flow to maintain riffle organisms

Invertebrate Sample Information

Qualitative Multi-Habitat Sample (QMH)

Divide 20 samples equally among habitat types present in the reach. If three habitat types are present take 7 samples in each of the three dominant habitats (for a total of 21). If a habitat is present, but not in abundance to sample in equal proportion to other habitats, sample as much as possible and divide the remaining samples between the dominant habitat types.

	Habitat			#
c	rock riffle/run rock substrate	Flow adequate to carry insects into net Artificial flow needed to carry insect		
c	aquatic macrophyte			
c	undercut bank, overhanging veg			
c	snag, woody debris, root wad			
c	leaf pack			

Number of multihabitat containers: _____

Additional Biological Information

Presence of freshwater sponge ---------- yes / no

Presence of exotic species ---------- yes / no

Name of exotic(s) if present:

(voucher a specimen if not present in sample)

Presence of mussels ----------yes / no

Description of mussel density and/or mussel bed location:

Notes

Pictures #: __ DD __ DU __ MD __ MU __ UD __ UU

Stream Sample External Label

MPCA Bioassessment – Invertebrate Sample

Sample Preservative - 100% reagent alcohol / 10% formalin

Sample Type: QMH / RTH

Sample Composition: Riffle / Bank / Wood / Veg

Date ____/____/20___ (mm/dd/yyyy)

Station Name _____

Station ID _____

Site Visit 1 / 2 Sample Jar ___ of ___

Collectors _____

Stream Sample Internal Label

Invertebrate Sample – sample type _____

Site Name:_____

Field Number_____

Date:_____/_____/_____ Bottle No. _____of____

Collected by: _____

_____