

## Section A1- Title Page

**DEPARTMENT OF WATER AND WASTE MANAGEMENT**

**DIVISION OF WATER AND WASTE MANAGEMENT**

**QUALITY ASSURANCE PROJECT PLAN**

**for**

**WATERSHED BRANCH MONITORING ACTIVITIES**

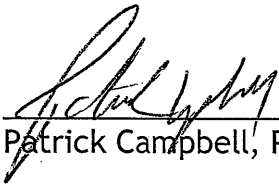


west virginia department of environmental protection

# Approval Sheet

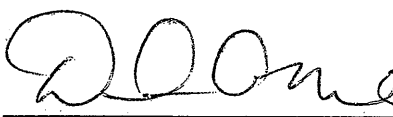
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Leo Essenthier, EPA Project Manager Date

  
Patrick Campbell, Project Manager 3/20/06  
Date

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John Wirts, Watershed Assessment Project Manager Date

  
Dave Montali, TMDL Project Manager 3/20/06  
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Janice Smithson, Project QA Manager 3/20/06  
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Dan Arnold, QA Officer Date

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# Chapter A: Project Management

## Section A3 Distribution List

This document and all supporting materials will be submitted to the following individuals. Distribution format will be electronic and/or paper copies.

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James Adkins, Environmental Resources Specialist II  
Jeffery Bailey, Wildlife Biologist II  
Andrew Johnson, Wildlife Biologist I  
Sydney Burke, Wildlife Biologist I  
Jason Morgan, Wildlife Biologist I  
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Michael Ong, Environmental Resources Specialist I  
Michael Puckett, Environmental Resources Specialist II  
Robert Row, Environmental Resources Specialist I  
Kevin Seagle, Environmental Resources Specialist I  
Kimberly Smith, Environmental Resources Specialist I  
Michael Whitman, Wildlife Biologist I  
Catherine Rosfjord, Environmental Resources Specialist I  
Nick Murray, Environmental Resources Specialist I  
Doug Wood, Environmental Resources Specialist III  
Michael McDaniel, Technical Analyst I  
Benjamin Lowman, Environmental Resources Specialist III

James Laine, Environmental Resources Specialist Supervisor  
Christine Daugherty, Environmental Resources Specialist III  
Judy Lyons, Environmental Resources Specialist I  
Stephen Stutler, Environmental Resources Specialist III  
Steve Young, Public Information Specialist  
James Summers, Environmental Resource Specialist III

## Section A4 Project/Task Organization

**Daniel Arnold, Quality Assurance Officer:** Supervises the Laboratory Certification Program; serves as Quality Assurance Officer for the activities of the Watershed Branch.

The Watershed Branch consists of the following personnel. An organization chart depicting the relationships of these individuals is presented in Figures 1A and 1B.

**Patrick Campbell, Assistant Director:** Supervises the Watershed Branch; responsible for program operations including budget; goals development and interagency coordination.

**Karen Light, Secretary I:** Assists in preparing budget; prepares invoices for payment; prepares monthly reports on expenditures; and assists staff in various projects.

**John Wirts, Environmental Resources Program Manager:** Supervises Watershed Assessment Section (WAS); responsible for program operations including budget, goals development, and intra-agency coordination.

**Janice Smithson, Wildlife Biologist III:** Supervises Biological Assessment Section; serves as field and biological QA/QC officer; prepares sampling lists and schedules for field crews; macroinvertebrate taxonomist; coordinates macroinvertebrate identification contract; Field Data Manager; prepares and maintains Quality Assurance Project Plan.

**James Adkins, Environmental Resources Specialist II:** STORET Data Manager; enters data into STORET; prepares GIS-based documents; performs data retrieval and data evaluations.

**Jeffery Bailey, Wildlife Biologist II:** Collects WAS and TMDL field data; macroinvertebrate taxonomist; trains and supervises summer interns; serves as Field Operations Manager and Biological Data Manager; performs biological data entry and assessment; prepares reports; co-leader of probabilistic sampling

project; co-leader of reference site criteria project; leader in Genus Level Index of Most Probable Stream Status (GLIMPSS) development; conducts site-specific benthic surveys.

**Andrew Johnson, Wildlife Biologist I:** Collects WAS and TMDL field data; conducts site-specific benthic surveys; lead periphyton biologist; participates in preparation of watershed reports.

**Sydney Burke, Wildlife Biologist I:** Collects WAS and TMDL field data; conducts site-specific benthic surveys; participates in the preparation of watershed reports; involved in permit-related studies; reviews field blank data and reports potential sampling errors; assists in QA/QC efforts; lead biologist for lake assessments; assists with periphyton assessments.

**Jason Morgan, Wildlife Biologist I:** Collects WAS and TMDL field data; macroinvertebrate and fish taxonomist; performs benthic data entry and macroinvertebrate QA/QC; conducts site-specific benthic surveys; involved in permit-related studies; involved with reference site selection and evaluation; lead in fish population and tissue collection studies.

**Karen Maes, Chemist I:** Data Entry Manager; performs and oversees data entry/review; manages and orders sampling supplies; assists in maintenance of benthic voucher and reference collections.

**Michael Arcuri, Environmental Resources Specialist Supervisor:** Supervises Water Quality Assessment Section; assists in planning WAS activities; oversees lake water quality monitoring. Staff is responsible for managing ambient water quality monitoring program, preparing watershed assessment reports, assisting with stream designated use assessments for 305(b) report, and maintaining program databases.

**Michael Ong, Environmental Resources Specialist I:** Collects WAS and TMDL field data; performs data entry and review; oversees monthly reporting and routine maintenance of state-owned field vehicles.

**Michael Puckett, Environmental Resources Specialist II:** Collects WAS and TMDL field data; assists with WCMS land-use characterization and checking lat./lon. Coordinates; performs data entry and review.

**Robert Row, Environmental Resources Specialist I:** Collects WAS and TMDL field data; performs data entry and review; oversees implementation of wetland monitoring strategy; oversees maintenance of boats and equipment; responds to public requests for water quality data; assists with fish population and tissue surveys; oversees diagnostics and minor repairs for in-situ water quality measurement instruments.

**Kevin Seagle, Environmental Resources Specialist I:** Oversees ambient water quality monitoring program; oversees deployment of remote water quality monitoring instruments; oversees special water quality studies.

**Kimberly Smith, Environmental Resources Specialist I:** Collects WAS and TMDL field data; prepares maps and tables for watershed assessment reports; performs stream designated use assessments for 305(b) reports; responds to public requests for water quality data; assists with Project Wet educational activities.

**Michael Whitman, Wildlife Biologist I:** Collects WAS and TMDL field data; Curator of voucher and reference macroinvertebrate collections; WAPBASE Data Manager; assists in maintenance, correction, and updating of WAS database; oversees integration of WCMS data and checking of coordinates in WAPBASE; Provides STORET and EQUiS database assistance; provides technical support to WAS staff, EPA, and co-operating agencies with database, GIS and other issues.

**Catherine Rosfjord, Environmental Resources Specialist I:** Collects WAS and TMDL field data; performs general data entry and review; assists with deployable water quality monitoring activities.

**Nick Murray, Environmental Resources Specialist I:** Collects WAS and TMDL field data; performs general data entry and review; provides assistance with public education activities.

**Doug Wood, Environmental Resources Specialist III:** Oversees preparation of watershed reports; assists with WAS and TMDL field sampling; member of DEP dive team.

**David Montali, Technical Analyst IV:** Serves as TMDL Program manager; supervises TMDL Development program and oversees preparation of WV Section 303(d) lists; responsible for program operations including goals development; planning and interagency coordination.

**Michael McDaniel, Technical Analyst I:** Coordinates the development of TMDLs for the state's water quality limited streams; develop contract documents and manages and oversees the contractual TMDL development process.

**Benjamin Lowman, Environmental Resources Specialist III:** Coordinates the development of TMDLs for the State's water quality limited streams; serves as staff specialist for biological impairment TMDLs and causative stressor identification.

**James Laine, Environmental Resources Specialist Supervisor:** Oversees the technical support and public outreach functions associated with TMDL development and 303(d) listing. Areas of responsibility include data management and compilation, pollutant source tracking, and public outreach.

**Christine Daugherty, Environmental Resources Specialist III:** General Data Manager and GIS Data Manager; oversees data management (Decision Database, Assessment Database, National Hydrological Dataset) and GIS needs of the Watershed Branch; updates DWWM website; assists in performing water quality data analysis; assists in preparation of 305(b) reports and 303(d) lists.

**Judy Lyons, Environmental Resources Specialist I:** Primary contact for Watershed Branch data requests; facilitates public meetings; gathers information from permit holders for TMDL development; prepares educational packets and brochures.

**Stephen Stutler, Environmental Resources Specialist III:** Collects and compiles data on waterbodies for use in preparing 303(d) lists and TMDLs; assists contractors and modelers in preparing TMDLs; and compiles permit data for mining operations for use in TMDL development.

**Steve Young, Public Information Specialist:** Public outreach coordinator; facilitates public meetings and hearings; conducts public outreach and educational campaigns; prepares Watershed Branch educational materials; assists in public outreach for DWWM; updates DWWM webpage.

**James Summers, Environmental Resource Specialist III (Working Title - Pollution Source Investigator):** Responsible for water pollution source tracking for TMDL program - collects water samples, field readings, field notes and GPS coordinates; maps sources in GIS format; coordinates exchange of information with WAS field samplers and TMDL modeling contractors.

Figure 1A. Watershed Branch Organization Chart - Watershed Assessment Section

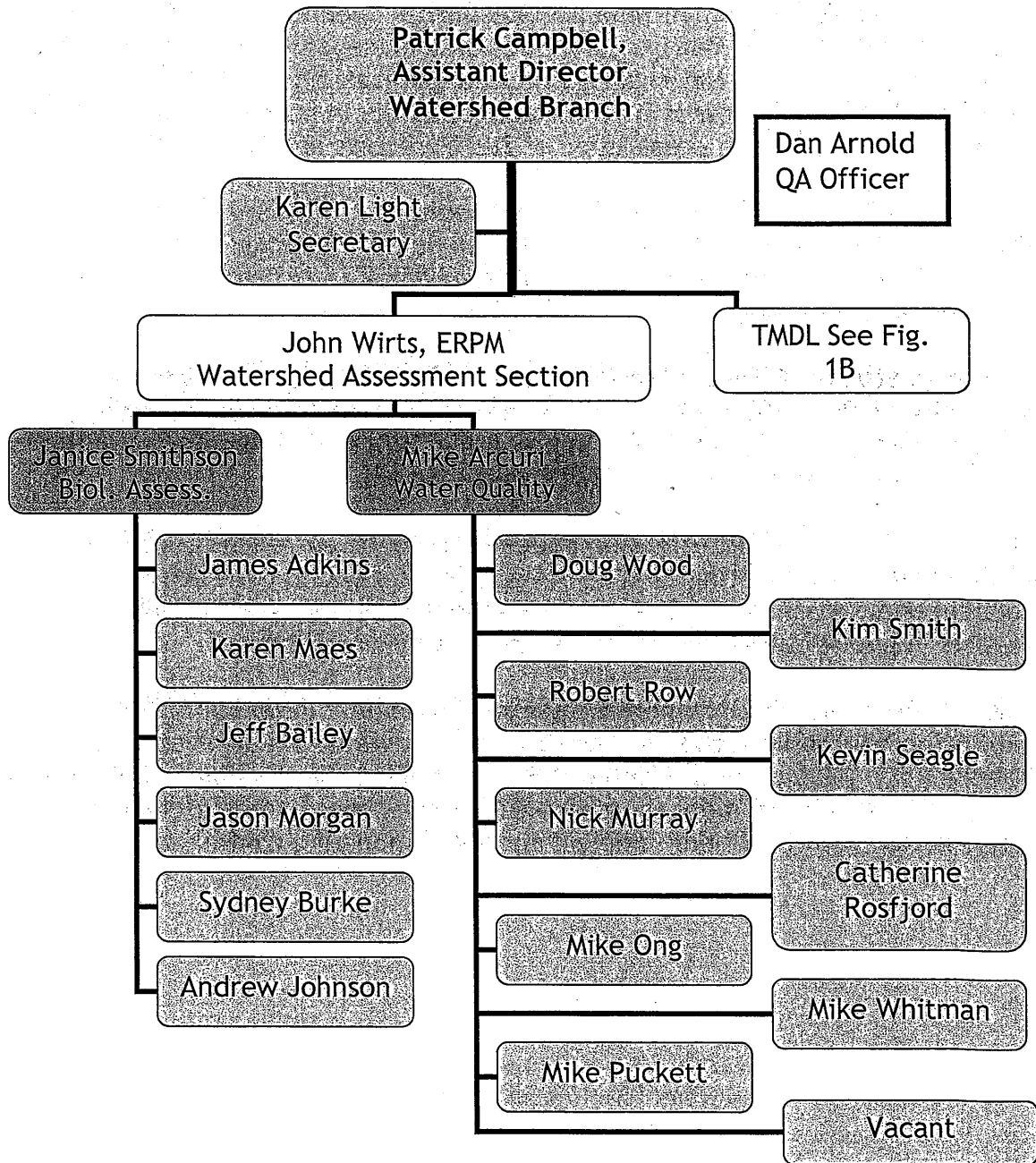
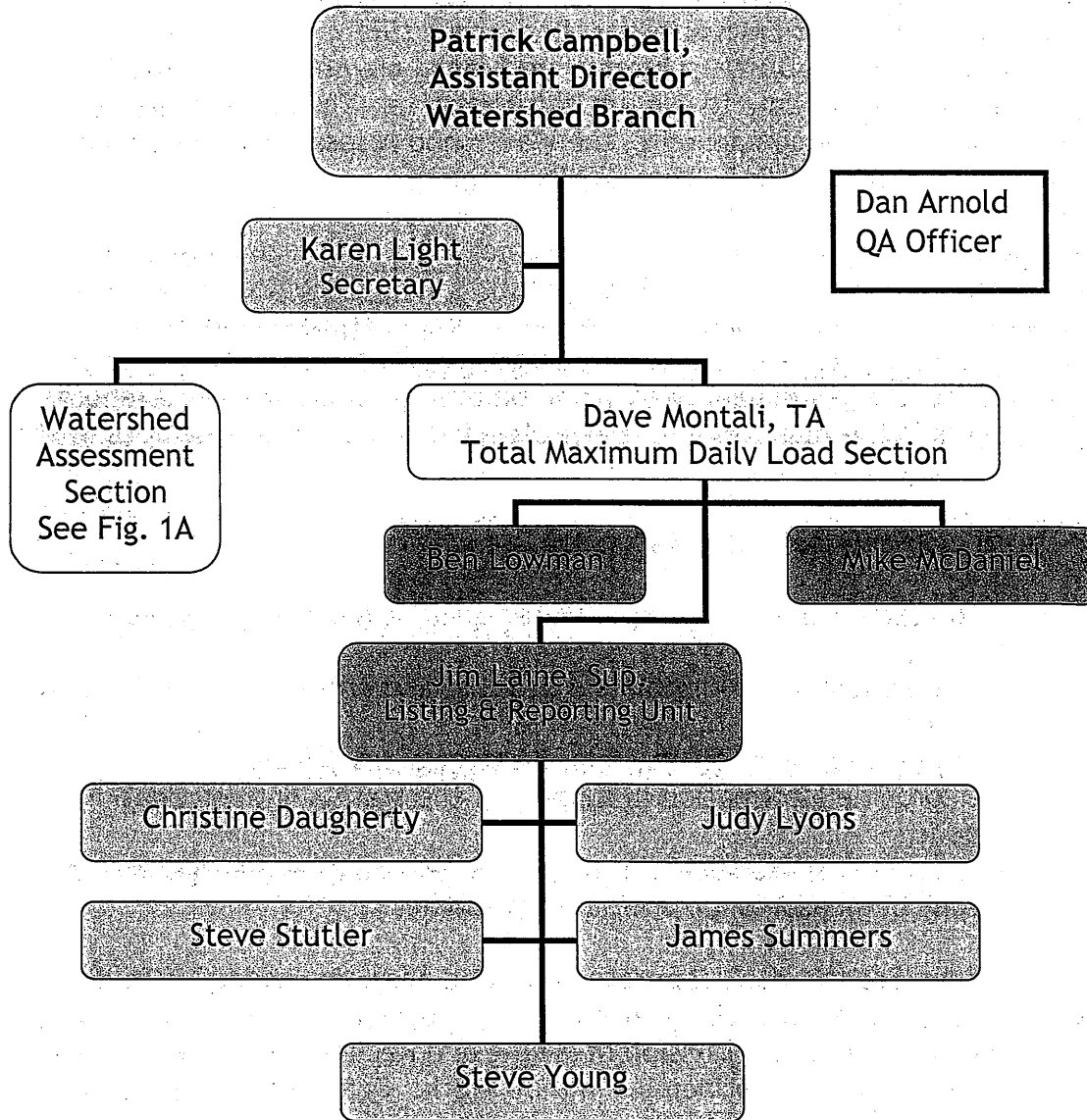


Figure 1B. Watershed Branch Organizational Chart - Total Maximum Daily Load Section.



## Section A5 Background and Problem Definition

The United States Environmental Protection Agency (EPA) promotes a watershed approach to monitoring, assessment and implementation of water quality protection activities. This approach provides an environmental management program that places a greater focus on ecosystems and utilizes decreasing resources more effectively in threatened watersheds. The West Virginia Department of Environmental Protection (DEP), Division of Water and Waste Management's (DWWM), Watershed Branch provides current water quality and biological data to support this initiative. The Watershed Branch consists of two major components, the Watershed Assessment Section (WAS) and the Total Maximum Daily Load (TMDL) Section.

The mission of the Watershed Branch is to collect and interpret water quality and biological information from West Virginia's 32 hydrological units on a five-year rotation. The data collected provide direction to stakeholders who regulate water quality and implement protective measures. As the five-year cycles repeat, the Watershed Branch will be able to measure the stakeholders' effectiveness in the management and protection of the water resources of the state.

The specific objectives of the Watershed Branch are:

- to obtain current, accurate water quality, habitat, and biological data;
- to maintain West Virginia's 303(d) list and prepare the state's 305(b) report;
- to prepare water quality improvement plans (TMDLs);
- to provide information in support of the state's antidegradation policy;
- to support stakeholders in the implementation of management and control measures for priority waterbodies.

The Watershed Branch also cooperates with and provides leadership to other DEP entities who are collecting water quality information. These entities (e.g., Non-point Source Program, Division of Mining and Reclamation, etc) often use Watershed Branch QA/QC principles in their work. Qualifying information is incorporated into the Branch's assessment and decision-making processes.



## Section A6 Task Description

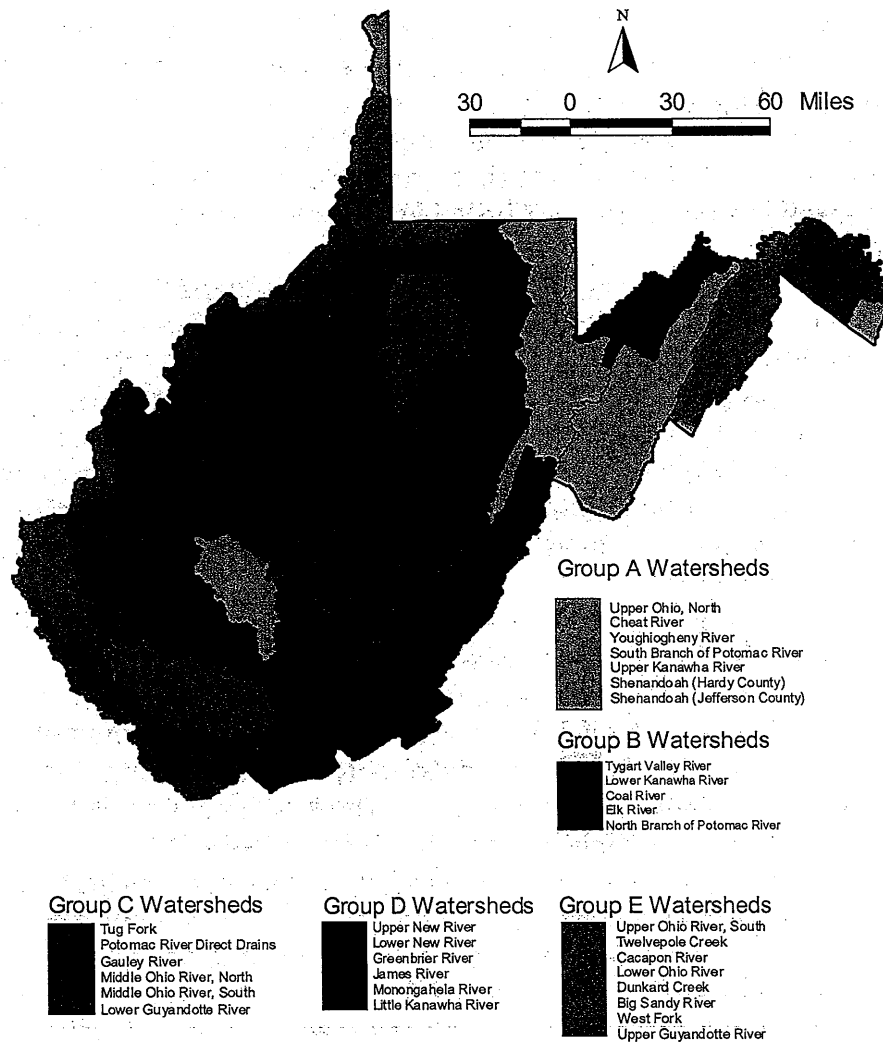
West Virginia has been divided in 32 waterbodies using the United States Geologic Survey's eight-digit cataloguing unit method. The cataloging units are as follows:

Code #	Watershed Name	HUC Code
1	South Branch of Potomac River	02070001
2	North Branch of Potomac River	02070002
3	Cacapon River	02070003
4	Potomac River Direct Drains	02070004
5	Shenandoah River (Hardy County)	02070006
6	Shenandoah River (Jefferson Co.)	02070007
7	James River	02080201
8	Tygart Valley River	05020001
9	West Fork	05020002
10	Monongahela River Direct Drains	05020003
11	Cheat River	05020004
12	Dunkard Creek	05020005
13	Youghiogheny River	05020006
14	Upper Ohio River, North	05030101
15	Upper Ohio River, South	05030106
16	Middle Ohio River, North	05030201
17	Middle Ohio River, South	05030202
18	Little Kanawha River	05030203
19	Upper New River	05050002
20	Greenbrier River	05050003
21	Lower New River	05050004
22	Gauley River	05050005
23	Upper Kanawha River	05050006
24	Elk River	05050007
25	Lower Kanawha River	05050008
26	Coal River	05050009
27	Upper Guyandotte River	05070101
28	Lower Guyandotte River	05070102
29	Tug Fork	05070201
30	Big Sandy River	05070204
31	Lower Ohio River	05090101
32	Twelvepole Creek	05090102

Each year, the Watershed Branch's data collection efforts will focus on four to eight of these watersheds. Upon the completion of a five-year cycle, each watershed will have been studied and the cycle is then repeated. The watershed sequence is presented below and is illustrated in Figure 2.

<b>Group A:</b>	<b>Assessment Years</b>
Upper Ohio River, North	2001
Cheat River	2006
Youghiogheny River	2011
South Branch of Potomac River	
Shenandoah River (Hardy County)	
Shenandoah River (Jefferson County)	
Upper Kanawha River	
<b>Group B:</b>	<b>Assessment Years</b>
Tygart Valley River	2002
Lower Kanawha River	2007
Elk River	2012
Coal River	
North Branch of Potomac River	
<b>Group C:</b>	<b>Assessment Years</b>
Middle Ohio River, North	2003
Middle Ohio River, South	2008
Potomac River Direct Drains	2013
Tug Fork	
Gauley River	
Lower Guyandotte River	
<b>Group D:</b>	<b>Assessment Years</b>
Lower New River	2004
Upper New River	2009
Greenbrier River	2014
James River	
Monongahela River Direct Drains	
Little Kanawha River	
<b>Group E:</b>	<b>Assessment Years</b>
Upper Guyandotte River	2005
Twelvepole Creek	2010
Upper Ohio River, South	2015
Cacapon River	
Lower Ohio River	
Dunkard Creek	
Big Sandy River	
West Fork	

Figure 2. Watershed Groupings for Assessment within the Five-Year Cycle.



The work performed by the Watershed Branch falls into several major categories: Watershed assessments, probabilistic sampling, TMDL development sampling, and the Ambient Water Quality Network (large river monitoring). These components are discussed in detail in the following paragraphs. Table 1 summarizes the biological, physical, and chemical interests for each of these activities. Figure 3 presents the timeline for various components.

**Watershed assessments** are synchronized with the five-year watershed cycle. Thus far, sampling efforts have emphasized on wadeable streams. Assessment sites are selected to address specific issues:

- **Reference sites:** These are unimpaired sites that must meet a specified set of criteria. A subset of previously identified reference sites is revisited in subsequent years and potential new reference sites will be considered for each watershed.
- **Impaired Streams:** Streams that are 303(d) listed or have impaired biological communities will be revisited and examined in greater detail.
- **“Gray” Streams:** These are streams having biological scores that fall inside an area of statistical uncertainty - the “gray zone”. Initial assessments at these sites failed to identify definitive impairment. Sites will be established to further characterize these streams.
- **Potential Tier 2.5 Streams:** Tier 2.5 is a subset of streams that contains naturally reproducing trout streams, reference streams, and streams having high biological scores. The Watershed Branch actively seeks to expand this list by selecting previously unassessed streams on public lands.
- **Unassessed streams:** Significant tributaries that have not been assessed during previous cycles.
- **Flood or Drought Impaired Streams:** This subset contains streams that were not in their normal state during previous assessment cycles. Scour from floods and extremely low flows temporarily depress the biological communities. These sites are revisited to evaluate their condition during normal flow conditions.
- **Significant Tributaries:** The watershed mainstem and significant sub-watersheds are sampled at multiple locations to examine the overall condition of the aquatic system and to determine spatial trends.
- **Small Streams with Low Biological Scores:** These streams scored low during previous assessments, but the causes of impairment are unknown. Resampling will help determine if natural conditions or anthropomorphic impacts are influencing the biological community.
- **Stakeholder Requests:** Specific requests from watershed associations and state and federal agencies are included during the site selection process.

Table 1. Summary of Watershed Branch Assessment Parameters.

Parameter	Sampling Activity			
	Probabilistic	Watershed Assess.	TMDL Devel.	Ambient
Habitat Evaluation	S	S	V	V
Physical Evaluation	S	S	S	S
Benthic Macroinvertebrates	S	S	V	V
Periphyton	S	S	V	V
Fish	V	V	V	
Stream Velocity			V	S
Field pH	S	S	S	S
Field Temp	S	S	S	S
Field Conductivity	S	S	S	S
Field Dissolved Oxygen	S	S	S	S
Hot Acidity	S	V	V	S
Cold Acidity		V		
Alkalinity	S	V	V	S
Hardness	S			S
Sulfate	S	V	V	S
Chloride	S	V	V	S
Fecal Coli.	S	S		S
Total Susp. Solids	S	V	V	S
Total Phosphate	S	V	V	S
Total Ortho-Phosphate			V	
Diss. Ortho-Phosphate			V	
TKN	S	V	V	S
Ammonia-N		V		S
Nitrate-Nitrite-N	S	V	V	S
Magnesium	S			
Manganese	S	V	V	S
Total Aluminum	S	V	V	S
Dissolved Aluminum	S	V	V	S
Dissolved Cadmium				S
Dissolved Copper	S			S
Total Iron	S	V	V	S
Dissolved Iron	S	V	V	S
Dissolved Lead				S
Total Calcium	S			
Total Selenium	S		V	S
Dissolved Selenium	S			
Total Arsenic				S
Dissolved Silver				S
Total Mercury	S			S
Dissolved Zinc	S			S
Dissolved Nickel				S
Volatile Organics				V
Semi volatile Organics				V

S=Standard Parameter collected during each event

V=Variable collected only as specified

The number of sites per year will vary depending on watershed size, level of effort per station, and personnel availability. Currently, sampling occurs April through October, inclusive. However, new research, based on data from previous watershed assessments, indicates that the benthic macroinvertebrate sampling period may be expanded to December through May and July through October.

During the initial cycle of watershed assessments (1996-2000), sampling efforts focused primarily on habitat, benthic macroinvertebrates, and water chemistry in wadeable streams. New components added during the second cycle (2000-2005) included periphyton collection and enhanced habitat evaluations. The Watershed Branch is planning to expand its efforts in the third cycle to include lakes assessments, non-wadeable streams and fish community components.

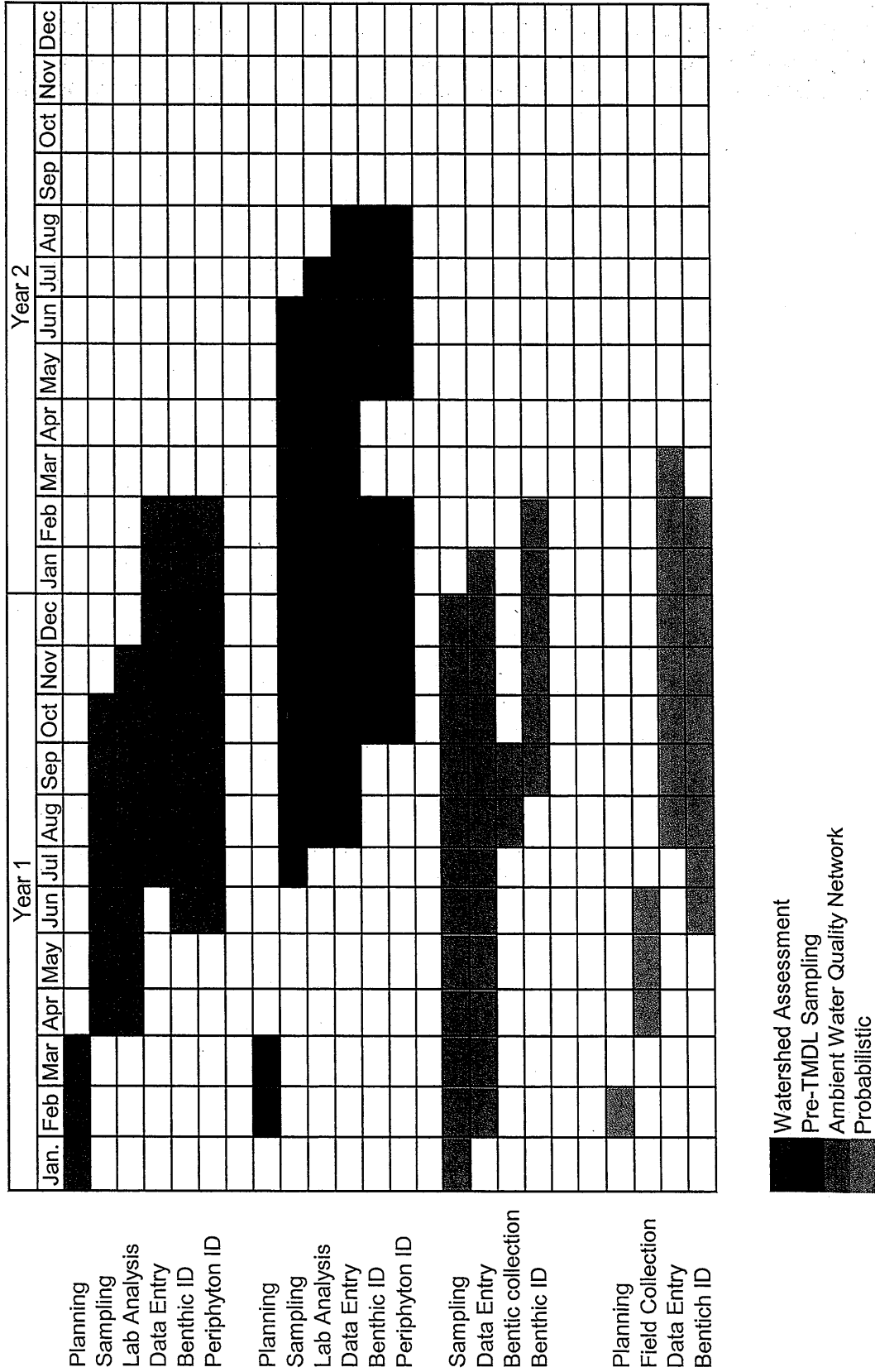
**Probabilistic sampling** produces unbiased data that can be subjected to statistical analysis with a high degree of confidence. These studies can be used to address state-wide issues, such as the biological differences among ecoregions or the number of stream miles impaired by sedimentation. Sites are selected at random using a program that can weight the site selection based on specified criteria, such as stream size, specific waterbody type, or ecoregion. The number of sites will vary depending on the objective of the study, but sampling efforts are sufficient to assure the data will stand up to statistical analysis. Typically, these studies have a duration of five years. However, probabilistic studies do not adhere to the five-year watershed cycle; instead, sampling occurs statewide annually. This process will help to mitigate problems that arise if a short-term environmental event, such as drought, occurs during the study.

**TMDL development sampling** is an intensive approach to obtaining a large amount of water quality information under a variety of environmental conditions. Conforming to the five-year cycle, sites are established on 303(d) listed streams and other streams that may provide additional supportive information. Water quality sampling is performed monthly, July through June. In addition, a subset of sites is subjected to a one-time biological assessment, which is performed following procedures established for watershed assessments. Data are submitted to TMDL modelers for development of pollution reduction plans.

**Ambient water quality monitoring** is performed to capture data from the state's larger rivers and streams. West Virginia's ambient monitoring network has been in existence since the mid 1940's, although the number of sites and sampling frequency has varied over the years. The current network consists of 25 fixed stations (Figure 4). Currently, these stations are visited quarterly; however, in 2006 bi-monthly sampling (i.e., six events per year) will be implemented. Each event includes brief documentation of prevailing

conditions and a large suite of water quality parameters. Additionally, these sites will be subjected to benthic macroinvertebrate sampling in August or September. Streams having wadeable riffles will be subjected to the same sampling protocols established for watershed assessments. Benthos are collected from non-wadeable streams using Hester-Dendy artificial substrates.

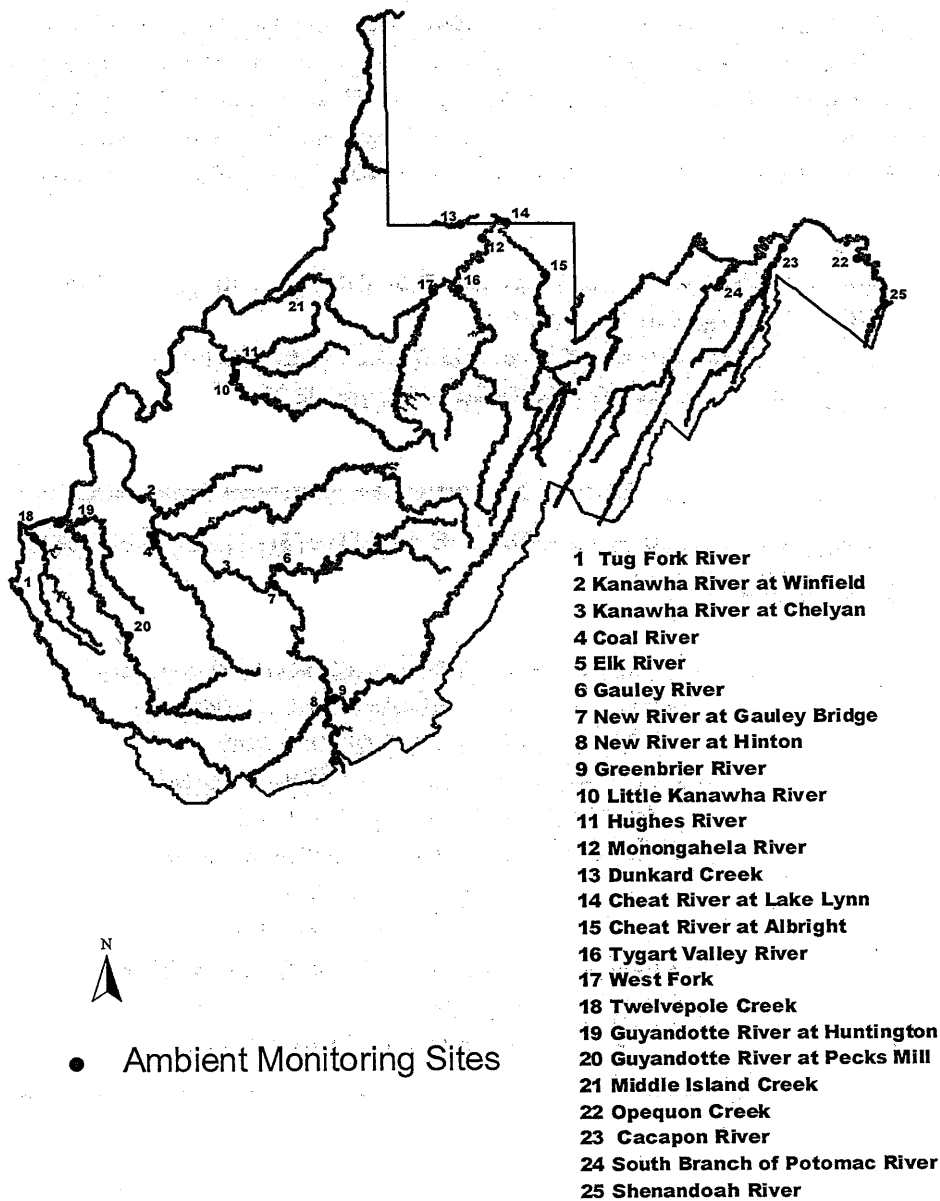
Figure 3. Annualized Timeline for Watershed Branch Activities.



Watershed Assessment  
 Pre-TMDL Sampling  
 Ambient Water Quality Network  
 Probabilistic



Figure 4. Map of Ambient Water Quality Network.



## Section A7 Quality Objectives and Criteria for Measurement Data

### Introduction

In 1996, the WV Office of Water Resources (now the Division of Water and Waste Management) initiated a new approach to address water quality issues by developing a statewide Watershed Management Framework. The objective of the watershed management scheme was to coordinate the operations of existing water quality programs and activities in West Virginia to achieve shared water resource management goals. On May 29, 1997, eleven agency and program directors from state and federal water quality agencies (Table 2) signed a resolution of mutual intent to form a partnership for statewide watershed management. The goals of the watershed management partners were to:

- ▣ Improve public awareness, understanding, and involvement
- ▣ Improve program efficiency
- ▣ Improve program effectiveness and cost effectiveness
- ▣ Improve information and data management

#### Table 2. Signature Agencies for the Partnership for Statewide Watershed Management

West Virginia Division of Environmental Protection  
West Virginia Soil Conservation Agency  
West Virginia Division of Forestry  
West Virginia Bureau for Public Health  
West Virginia Bureau of Commerce  
U. S. Environmental Protection Agency  
U. S. Geological Survey  
U. S. Office of Surface Mining  
U. S. Forest Service, Monongahela National Forest  
Natural Resource Conservation Service  
U. S. Army Corps of Engineers

The five phases of the Watershed Management Framework are as follows:

1. Scoping and Screening - compile existing data and conduct public outreach to identify problems and issues within watersheds.
2. Strategic Monitoring and Assessment - develop and implement a monitoring plan and conduct monitoring assessments.
3. Management Strategy Development - develop and assess integrated management strategies, including the development of TMDLs.
4. Priority Watershed Management Plan - develop and finalize management plans.
5. Implementation - implement point and non-point management strategies.

## Quality Objectives

The West Virginia Department of Environmental Protection created the Watershed Assessment Program (currently known as the Watershed Branch) to help address the needs of these stakeholders by implementing Phases 2 and 3 of the framework. The data quality objective process the Watershed Branch uses to address the needs of the stakeholders is presented in Appendix A.

## Criteria for Measurement Data

All water quality samples are tested at West Virginia certified laboratories. Certification assures that Data Quality Indicators<sup>1</sup> (DQIs) are in compliance with quality assurance/quality control protocols. Analytical methods are specified in 40 CFR 136. Lab blanks, spiked samples and duplicates must be performed at specified intervals. Detection limits must fall below the action level.

Standardized methods are used for habitat evaluations and the collection of water samples and biological assemblages. A series of Standard Operating Procedure (SOP) manuals detail field protocols. All new personnel are subjected to intensive training with seasoned biologists. Duplicate sampling and field blank preparation are required.

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<sup>1</sup> Data quality indicators are performance and acceptance criteria. Examples of DQIs are precision, bias, accuracy, comparability, and sensitivity.

## Section A8 Special Training and Certification

All field personnel must participate in an annual training session. These sessions are designed to provide hands-on experience with field protocols. Topics include all aspects of data collection; however, the emphasis is placed on newly implemented procedures and components that can be influenced by personal bias, such as habitat evaluations. Trainers are also part of the field crew and, as such, can provide ongoing evaluations of teammates throughout the field season.

Safety is an issue that cannot be ignored. Field personnel receive first aid and CPR training periodically. All personnel involved in the collection of fish through electro-shocking must be aware of all safety precautions and all power-generating equipment must have functioning emergency shut-off switches. Biologists in charge of the fish collection program have successfully completed the U.S. Fish and Wildlife Service National Conservation Training Center's electrofishing course. Safe boating courses are also available.

## Section A9 Documents and Records

Signatory personnel will receive paper copies of this Quality Assurance Project Plan. Other recipients identified in the Distribution List (Section A3) will receive electronic copies via email. A copy of this document will also be available on a shared hard-drive.

Standard Operation Procedures are made available to all Watershed Branch personnel. In addition, all field personnel receive bound hard copies for reference during sampling events. SOPs are reviewed annually and are updated or supplemented as necessary.

Documentation of instrument calibration and repair is maintained in binders in the Watershed Branch laboratory at the DEP headquarters. Field forms, chain-of-custody forms, and analytical results are organized and filed. Reference material for biological assemblages is maintained in the laboratory.

All documents and records are maintained for a minimum of five years. Paper files are stored at the DEP headquarters. Anyone removing a file is required to sign it out so that it can be easily located. Older versions of Quality Assurance Project Plans and SOPs are retained as electronic files. All electronic data are backed up daily by DEP's Information and Technology Office.

# Chapter B: Data Generation and Acquisition

## Section B1 Sampling Process Design

This Quality Assurance Project Plan embodies the four primary monitoring components of the Watershed Branch: General watershed assessments, probabilistic sampling, pre-TMDL sampling, and ambient water quality monitoring. The sampling design process for each of these components is described in the following paragraphs. Specific elements of these projects are presented in Table 1.

### General Watershed Assessments

The objectives of general watershed assessments are to identify areas of impairment and to document recovery in areas where pollution abatement activities have been implemented. A directed sampling approach is employed for general assessments. Sites are selected to confirm and update existing data, or to address questions arising from previous assessments. Unassessed waterbodies may also be targeted. The study area is selected in accordance with the five-year cycle described in Section A6.

All sites are selected in advance, but field personnel have the freedom to move a site to obtain the best representative sample or if the designated site is inaccessible. The number of sites per year will vary, depending on the size of the watershed. Critical elements are habitat evaluations, macroinvertebrate and periphyton assemblages, on-site measurements (pH, dissolved oxygen, water temperature, and conductivity), and fecal coliform bacteria. Additional water quality parameters may be obtained at the discretion of the collectors. A set of reference sites serves a background population. Reference sites have minimal anthropogenic influence and must comply with a set of "high-quality" criteria.

This project is seasonal with sampling taking place April through October, inclusive. (New research, based on data from previous watershed assessments, indicates that dual seasons - December to May and July to October - may be more appropriate.) Samples are collected over a rather short time frame; usually a few weeks per watershed. These are single-sample events; however, a site may be revisited on subsequent assessment cycles. Water samples are mid-stream grabs; they are preserved according to regulation and retained by the sampler until laboratory pick-up. Benthic macroinvertebrate and

periphyton samples consist of composites. Replicates and field blanks are prepared at 4-5% of the sites. Replicate sampling is a complete duplication of effort; that is, teammates switch roles so that each person performs a complete assessment and collects all samples.

Rainfall events may cause delays in the schedule. High waters and turbidity obscure key components of habitat evaluations and prohibit the collection of biological samples.

## Probabilistic Sampling

The objective of probability sampling is to obtain data that can provide strong statistical conclusion. Depending on the specific objective of the probabilistic research, sampling may occur state-wide or be restricted to specific watersheds. Sites are randomly selected using the general protocols employed in EPA's R-EMAP Program<sup>2</sup> described in *R-EMAP: Regional Environmental Monitoring and Assessment Program* (EPA, 1993). This randomization process assures that no particular portion of the group of stream is favored; the chance of selecting a degraded site is proportional to the number of streams having degraded conditions. Results of probabilistic study designs can be used to characterize watersheds, ecoregions, or the entire state.

The prime objective for the current probabilistic study is to obtain sufficient data for both statewide and watershed-specific applications. This five-year study, initiated in 2002, entails annual statewide sampling. This design minimizes the effects of short term events, such as droughts, that may periodically impact an area. Six samples are collected from each watershed<sup>3</sup> annually to meet statistical requirements. This process will provide 30 samples per watershed at the conclusion of the study.

Sites are selected in advance and cannot be moved. If a site is inaccessible or unsamplable, it is replaced by a new randomly-selected site. The annual number of sites sampled is 150. Critical elements include habitat evaluations, macroinvertebrate and periphyton assemblages, on-site measurements (pH, dissolved oxygen, water temperature, and conductivity), and water quality sampling (Refer to Table 1 for details).

The one-time sampling events take place between April and June, inclusive. Water and biological samples are obtained and handled according to the same

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<sup>2</sup> R-EMAP: Regional Environmental Monitoring and Assessment Program. 1993. United States Environmental Protection Agency. EPA/625/R-93/012.

<sup>3</sup> Small watersheds (<30 streams) were combined with larger watersheds having similar characteristics.

protocols applied to general watershed assessments. Replicate sampling is conducted at 2% of the sites and field blanks are prepared weekly.

## Pre-TMDL Sampling

Large amounts of data from impaired streams are needed to develop accurate TMDL models. Therefore, a targeted sampling plan is used to develop pre-TMDL sampling networks. The objective of this sampling scheme is to obtain as much information on a watershed or sub-watershed as budget and personnel restrictions allow. Sites are selected in advance with these limitations in mind. TMDL planners consider all existing data in designing the network. Sites are selected based on known or suspected impairments. Additional sites are established upstream of known impacts and/or on nearby undegraded streams to provide information on background conditions.

The TMDL network for any given year may consist of hundreds of sites. To manage field activities, the network is broken down into smaller groups of ~50 sites, based on travel logistics. Each "mini-network" is permanently assigned to an individual, who is responsible for data collection for the duration of the study.

Although sites are designated in advance, field personnel are permitted to move sites with the approval of the TMDL decision-makers. Critical elements are water quality samples (specific parameters will vary), on-site measurements, and documentation of field observations. Some sites may require one-time macroinvertebrate and periphyton collections as well.

Sampling is conducted monthly July through June. Networks are completed as quickly as possible, usually in one to two weeks. Grab water samples are preserved in accordance with established protocols and held until laboratory pick-up is arranged. Individuals assigned to the network are required to perform replicate sampling and prepare field blanks during each event. Sampling is conducted regardless of weather conditions; however, field crews are not required to work in unsafe conditions. Delays may occur in obtaining the biological components if scouring floods or droughts occur.

## Ambient Water Quality Network

The Ambient Water Quality Network was established to evaluate long-term spatial and temporal trends in the state's larger streams and rivers. This network of sites was established in the 1940's, but the number of sites and sampling frequency has varied over the years.

The current network consists of 25 sites that are sampled bi-monthly (six times per year). Critical elements included on-site measurements, field observations, and water quality parameters (Refer to Table 1). Macroinvertebrate communities will also be sampled in late summer/early fall.

A single individual is responsible for the Ambient Network and sample collection is scheduled around his/her other activities. Water samples are mid-stream or streambank surface grabs. Samples are appropriately preserved and held until laboratory pick-up can be arranged. One replicate sample and one field blank are prepared each month or a rate of 8%. Trip blanks for organic samples are prepared daily.

## Section B2 Sampling Methods

Detailed descriptions of sampling protocols, instruments, sampling devices, sample containers, forms, and guidance are contained in the Watershed Branch SOPs (Appendix B). All activities covered in this quality assurance project plan are addressed in these documents. Annual training sessions and adherence to these SOPs assures that the data generated by the Watershed Branch are comparable and defensible.

In any endeavor things can - and will - go wrong. Fortunately, field crews rarely work in isolation. If one team experiences an equipment failure or supply shortage, a team working in an adjacent area can offer support or advice on possible solutions. Replacement supplies, such as sample containers, can be provided by the lab during sample pick-up. In some cases, arrangements can be made with field offices or headquarters for supply or equipment replacement. If a sample is lost or destroyed after collection, the site may be revisited as long as conditions are comparable to the original situation. Replacement samples must be clearly identified.



## Section B3 Sample Handling and Custody

Sample handling and preservation for all samples, except fecal coliform bacteria, conforms to methods specified in 40CFR136. These methods are detailed in the specific SOPs and are summarized in Table 3. All samples are collected in sufficient quantities to perform analyses with enough excess for duplicate analysis.

Holding times for fecal coliform bacteria samples were expanded to 24 hours. Sampling can occur in remote areas and the 6 hour holding time was impractical or impossible to attain. The Watershed Branch conducted literature searches and performed internal testing to discern the differences in fecal colony counts based on 6-hour and 24-hour holding times. It was determined that decision errors based on 24-hour fecal samples would be insignificant.

Table 3. Sample Collection and Preservation Methods.

Parameter	Container	Preservation Method	Maximum Holding Time
Benthic Macroinvertebrates	Wide mouth plastic jar	75% Denatured Ethanol	Indefinite
Periphyton	4 oz specimen jar	10% formalin, Cool <4°	Indefinite
Hot Acidity	1-L Cubitainer	Cool, 4°C	14 days
Cold Acidity	1-L Cubitainer	Cool, 4°C	14 days
Alkalinity	1-L Cubitainer	Cool, 4°C	14 days
Hardness	1-L Cubitainer	Calculated value	
Sulfate	1-L Cubitainer	Cool, 4°C	28 days
Chloride	1-L Cubitainer	None	28 days
Fecal Coliform Bacteria	100 ml sterile bottle	Cool, 4°C, +sodium thiosulfate	24 hours*
Total Susp. Solids	1-L Cubitainer	Cool, 4°C	7 days
Total Phosphate	1-L Cubitainer	Cool, 4°C, H <sub>2</sub> SO <sub>4</sub> to pH<2	28 days
Total Ortho-Phosphate	1-L Cubitainer	Cool, 4°C	48 hours
Diss. Ortho-Phosphate	1-L Cubitainer	Filter immediately, Cool, 4°C	48 hours
TKN	1-L Cubitainer	Cool, 4°C, H <sub>2</sub> SO <sub>4</sub> to pH<2	28 days
Ammonia-N	1-L Cubitainer	Cool, 4°C, H <sub>2</sub> SO <sub>4</sub> to pH<3	28 days
Nitrate-Nitrite-N	1-L Cubitainer	Cool, 4°C, H <sub>2</sub> SO <sub>4</sub> to pH<4	28 days
Magnesium	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Manganese	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Aluminum	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Aluminum	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Cadmium	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Copper	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Iron	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Iron	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Lead	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Calcium	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Selenium	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Selenium	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Arsenic	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Silver	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Total Mercury	1-L Cubitainer	HNO <sub>3</sub> to pH<2	28 days
Dissolved Zinc	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Dissolved Nickel	1-L Cubitainer	HNO <sub>3</sub> to pH<2	6 months
Volatile Organics	40-mL Glass Vial	HCl to pH<2	14 days
Semivolatile Organics	1-L Amber Glass Jar	None	7 days

\* This holding time is not in compliance with Standard Methods.

All samples are labeled to indicate the station code, waterbody name, date/time of collection, and type of preservative. An "Analysis Request Form" is used to indicate the desired analyses. The bottom of the form contains a section to track sample chain-of-custody. Examples of sample labels and forms are included in the SOPs.

Labs are required to document sample receipt and assign tracking numbers. After the holding time has expired, the receiving lab properly disposes the samples.

## Section B4 Analytical Methods

All water samples submitted to laboratories are analyzed in accordance with 40CFR136 or SW-846. Protocols for field measurements and benthic samples are defined in the SOPs (Appendix B). Periphyton samples are processed according to specifications defined in the contract (Appendix C).

## Section B5 Quality Control

Multiprobe instruments are used to determine pH, dissolved oxygen, conductivity, and temperature. These units are fully calibrated weekly, prior to sampling in the field. Calibration, adjustments, and maintenance are recorded for each instrument. Any instrument failing to meet calibration requirements is repaired on-site or returned to the manufacturer. All repairs are documented in the calibration books. The identification number for each unit is recorded each time the meter is used. This process allows minimum loss of data, if the meter fails in the field or upon recalibration.

Field crews are required to conduct replicate sampling and field blanks for each "list" of samples collected. All sampling efforts are broken down into smaller, more manageable components or "lists", which are then assigned to an individual or crew for completion. In order to complete a list, a replicate sample and field blank must be performed. The frequency of quality control activities will vary, depending on the length of the list. Estimated rates for the various projects are: Pre-TMDL - 3%, General Watershed Assessments - 5%, Probabilistic Sampling - 2.5%, and Ambient Water Quality Network - 7.6%.

The intensity of replication is dependant upon the specific project. General watershed assessments and probabilistic sampling is performed by two-person crews. A random number generator is used to assign specific sites for replication or field blank preparation. Field crews have the liberty to move a

replicate to a different site, if the assigned replicate site is unsuitable. At a replicate site, each crew member conducts a full assessment as though he/she is the only one present.

Pre-TMDL and Ambient Network sampling is usually performed by an individual working alone. Sites for replication and field blanks are determined by the list assignee. Only water quality sampling is replicated as replicate habitat observations would be redundant. When semi-volatile and volatile organics are collected for the Ambient Network, trip blanks are prepared daily.

Various steps may be taken if a field blank exceeds an analyte detection limit. If the analyte detected is near the analytical limits and is significantly lower than actual field conditions, it is considered an outlier and no additional action is taken. If a field blank is significantly higher than the minimum detection limit, steps are taken to verify that the field blank was not misidentified. The lab is notified of the situation and asked to verify the results. If it is clear that an error has been made and is correctable, the data are revised and the reasons fully-documented.

All water analyses are performed by firms that have been awarded contracts by the state. The specifications for this contract include quality control requirements. Quality control requirements are described in detail in the contract (Appendix C). Stated briefly, all labs are required to use standard analytical procedures. Duplicates and spikes must be performed every tenth sample and reference samples must be tested every six months. In addition, DEP may submit blind samples of known composition.

Benthic macroinvertebrate samples are processed and identified by contracted facilities. The current contract is presented in Appendix C. The contract specifies sample sorting and identification procedures by referencing the Watershed Branch SOPs. Quality control processes, provided in detail in the contract, require documentation of sorting efficiency, and internal identification accuracy. Voucher and reference collections are developed and returned to the Watershed Branch when the project is completed. A minimum of 2.5% of the returned samples are verified by a second laboratory. Failure to comply with comparability assessments may result in contract cancellation. Future contracts will also specify that the taxonomists have certification from the North American Benthological Society for genus-level identification.

Periphyton analysis is performed by contracted laboratories. The contract does not specify exact quality control procedures. However, the all bidders must present acceptable quality control measures to be considered for the contractual award. A copy of the contract is presented in Appendix C.

## Section B6 Instrument/Equipment Testing, Inspection, and Maintenance

Hydrolab and YSI brand multiprobe instruments are used to measure pH, dissolved oxygen, temperature, and conductivity in the field. Each instrument is individually numbered so that its history may be traced. Before use, the probes are examined for fractures, punctured membranes, biofouling and other problems. Probes are cleaned, repaired, or replaced as needed. Spare probes are available as replacement parts.

Flow measurement equipment is zero-adjusted annually. The probe is cleaned if readings become erratic. However, erratic readings may also occur in waterbodies with high conductivity or near-freezing temperatures.

Any instrument that cannot be repaired on-site is shipped to the manufacturer for repair. All repair and maintenance activities are recorded in the instrument's calibration manual.

Benthic macroinvertebrate sampling nets are examined for holes prior to each use. Worn nets are replaced as needed. After each use, the net is rinsed thoroughly and examined closely for any organisms that may be clinging to it.

One person is assigned responsibility for ordering and maintaining supplies. This individual monitors usage of sample bottles, batteries, preservatives, etc., and reorders these items as required. Field personnel are provided with checklists to assure that vehicles are fully stocked prior to departure.

## Section B7 Instrument/Equipment Calibration and Frequency

Instructions for calibrating all instruments are detailed in the SOPs. Hydrolab and YSI brand multiprobe instruments are calibrated weekly prior to use. Additionally, dissolved oxygen is calibrated daily as this parameter tends to drift with changes in elevation and barometric pressure. All calibration activities, including maintenance and repairs are recorded in the calibration manuals. Flow measurement probes are zeroed annually.

## Section B8 Inspection/Acceptance of Supplies and Consumables

Critical supplies include Cubitainer brand bottles (for general water samples), sterile fecal coliform bottles, macroinvertebrate and periphyton sample jars, field and chain-of-custody forms, preservatives (acids, formalin, alcohol), batteries, deionized water for rinsing and field blank preparation, and calibration standards.

The supply officer monitors the levels of these consumables and orders new supplies from the current state laboratory-supply contract. The supply officer examines these items when they are received and documents the receipt and expiration date for preservatives and standards. All consumables that exceed the expiration date are discarded.

## Section B9 Non-direct Measurements

The Watershed Branch relies on internally-collected data for most of its decision-making processes. However, data provided from outside sources - watershed associations, mining and permitting surveys, etc. - are also taken into consideration. These data are considered supportive; that is, they are used to help prove the assumptions made from the Watershed Branch's data. Greater weight is given to outside data that are known to be collected using Watershed Branch protocols. Any data that meet the strict requirements of the Watershed Branch and are known to be obtained using the Branch's protocols are entered into the Decision Database.

## Section B10 Data Management

All paper data - field forms, chain-of-custody forms and assignment lists - are submitted to the Field Data Manager. These documents are compared to the original assignment list and reviewed for completeness and reporting errors. Laboratory results, which are submitted in both electronic and paper formats, are collated with the respective field documents. The electronic version of the collated materials is then transferred to the Watershed Branch's database: WAPBASE.

WAPBASE uses the Microsoft Access program and readily imports data from other databases and/or spreadsheets. It is stored on DEP's mainframe and is backed-up daily.

After the electronic data have been merged into WAPBASE, the paper versions are submitted to the Data Entry Manager to key in additional information that could not be transferred electronically. The Data Entry Manager is responsible for assuring that all data have been entered and reviewed for transcription errors. The data entry and review processes are documented within the database. Errors that are noticed after the final review process are also documented within the database. The Data Entry Manager is also responsible for proper filing of paper copies in the central file room at DEP's headquarters.

The STORET Data Manager uses the WAPBASE data to enter Watershed Branch information into EPA's national repository for water quality data.

The WAPBASE Data Manager is responsible for overall maintenance of the database containing information collected in the field. This individual is responsible for WAPBASE setup, design, security, data transformation and reduction, and system backup. The General Data Manager also reviews data for consistent format and site-location errors. WAPBASE is backed-up daily by the DEP Information and Technology Office. The database is also backed-up on DVD before and after major revisions. The WAPBASE General Data Manager also maintains supportive databases: Taxonomic lists, a master stream list, and the Division of Natural Resources Stream Reach File.

The General Data Manager oversees databases regarding the decision-making and assessment processes. The Decision Database (DDB) imports data from WAPBASE. The DDB contains information required for 303(d) List preparation and records information required for making critical decisions, such information as the number of water quality violations. The DDB also houses information on specific impairments and TMDL development. Final decisions are then reported in the Assessment Database (ADB). The ADB is used to document impaired streams and to indicate whether they are fully supporting or non-supporting their designated uses. The ADB also indicates circumstances where data are insufficient to make fully/non-supporting decisions. If a stream is designated as non-supporting the ADB will list the causes and sources of impairment. The General Data Manager also oversees GIS-related databases and the National Hydrology Dataset.

Members of the Watershed Branch have read/write capabilities for WAPBASE. Others within DEP have access to WAPBASE as read-only status. This system allows many users to query WAPBASE for information.

DEP is developing a centralized system to incorporate all data the agency generates into a single database. This system, known as EQUiS, is still in the developmental stages and cannot be discussed at this point.

# Chapter C Assessment and Oversight

## Section C1 Assessments and Response Actions

The Watershed Branch conducts assessments of its activities to assure the requirements of the Quality Assurance Project Plan are being implemented. These assessments are discussed below and summarized in Table 4.



Table 4. Assessments and Response Actions

Assessment Type	Frequency	Internal or External	Organization Performing Assessment	Person, title, Organization Affiliation Responsible For:			Monitoring Effectiveness of Corrective Actions
				Performing Assessment	Responding to Assessment Findings	Identifying and Implementing Corrective Actions	
Readiness Review TMDL	Annually	Internal	WB/TMDLS	Dave Montali, TA, TMDLS Ben Lowman, ERS, TMDLS Michael McDaniel, TA, TMDLS	WB - Field Personnel	Michael McDaniel, TA, TMDLS Ben Lowman, ERS, TMDLS	Michael McDaniel, TA, TMDLS Ben Lowman, ERS, TMDLS
Readiness Review WAP	Annually	Internal	WB/WAS	Jeffery Bailey, WLB, BAP John Wirts, ERP, WAS	WB - Field Personnel	Jeffery Bailey, WLB, BAP John Wirts, ERP, WAS Janice Smithson, WLB, BAP Mike Arcuri, ERSS, WQAP	Jeffery Bailey, WLB, BAP John Wirts, ERP, WAS Janice Smithson, WLB, BAP Mike Arcuri, ERSS, WQAP
Surveillance Field	Continuous	Internal	WB/WAS	Jeffery Bailey, WLB, BAP Mike Arcuri, ERSS, WQAP Janice Smithson, WLB, BAP Sydney Burke, WLB, BAP	WB - Field Personnel	Jeffery Bailey, WLB, BAP Mike Arcuri, ERSS, WQAP John Wirts, ERP, WAS Janice Smithson, WLB, BAP	Jeffery Bailey, WLB, BAP Mike Arcuri, ERSS, WQAP Janice Smithson, WLB, BAP John Wirts, ERP, WAS
Surveillance Lab	Continuous	External	WB/WAS	Janice Smithson, WLB, BAP Karen Maes, CH, BAP	Contracted Laboratories	Contracted Laboratories	Janice Smithson Karen Maes, CH, BAP
Surveillance Data Mgmt	Continuous	Internal	WB/WAS	Janice Smithson, WLB, BAP Karen Maes, CH, BAP Mike Whitman, WLB, WQAP Christine Daugherty, ERS, TMDLS	Janice Smithson, WLB, BAP Karen Maes, CH, BAP Mike Whitman, WLB, WQAP Christine Daugherty, ERS, TMDLS	Janice Smithson, WLB, BAP Karen Maes, CH, BAP Mike Whitman, WLB, WQAP Christine Daugherty, ERS, TMDLS	Janice Smithson, WLB, BAP Karen Maes, CH, BAP Mike Whitman, WLB, WQAP Christine Daugherty, ERS, TMDLS
Proficiency Testing	Periodic	External	LQAP	Dan Arnold, QAO, LQAP Sydney Burke, WLB, BAP	Dan Arnold, QAO, LQAP	Dan Arnold, QAO, LQAP	Dan Arnold, QAO, LQAP
System Audits	Periodic	External	EPA	EPA Wheeling Biologists	WB - Personnel	EPA Wheeling Biologists John Wirts, ERP, WAS Janice Smithson, WLB, BAP Mike Arcuri, ERSS, WQAP	John Wirts, ERP, WAS Janice Smithson, WLB, BAP Mike Arcuri, ERSS, WQAP

WB=Watershed Branch of WV Department of Environmental Protection  
WAS=Watershed Assessment Section of the Watershed Branch  
TMDLS=TMDL Section of the Watershed Branch  
BAP=Biological Assessment Program of WAS  
WQAP=Water Quality Assessment Program of WAS  
LQAP=Laboratory Quality Assurance Program

ERP=Environmental Resource Program Manager  
ERS=Environmental Resource Specialist  
ERSS=Environmental Resource Specialist Supervisor  
WLB=Wildlife Biologist  
TA=Technical Analyst  
CH=Chemist, QAO=Quality Assurance Officer

Readiness reviews are conducted prior to the start of a new activity or at major milestones, such as beginning a new TMDL or Watershed Assessment Cycle. These reviews are typically conducted during the annual training event and staff meetings. The ability to conduct entirely new activities, such as fish population studies, is tested through the implementation of pilot studies.

Surveillance is a continuous process of verification; it assures that all activities are being performed to specification. For Watershed Branch purposes, surveillance may be broken down into three categories: Field, lab, and data management activities. The Field Operations Manager and/or supervisors in the Watershed Assessment Section spend time in the field with each team member to assure work is being performed in accordance with the SOPs. Surveillance of laboratory activities includes assuring that all data are received and meet minimum detection requirements. A team of data managers assures the accuracy of electronic information.

To evaluate the proficiency of water testing laboratories, a sample having known quantities is submitted to the testing facility as a blind sample. Results of proficiency testing must fall within specified acceptance criteria. This aspect of assessment is managed by the Laboratory Quality Assurance Program.

System audits are thorough systematic on-site assessments. Laboratory audits are a component of the Laboratory Quality Assurance Program. In order to be contracted for testing DEP samples, a laboratory must have successfully passed a system audit and must be certified by the state. The activities of the Watershed Branch are audited periodically by EPA biologists stationed in the Wheeling Field Office.

## Section C2 Reports to Management

West Virginia's Integrated Report, the Watershed Branch's Ecological Assessments and completed TMDLs are the primary vehicles for summarizing the methodologies and information collected on the state's waters. These important documents summarize and describe large volumes of data representing assessment and TMDL development expenditures approaching three million dollars a year.

Management of the activities necessary to produce these reports is an active and ongoing process. Monthly detailed intra-branch management reviews provide the systematic approach necessary to ensure that quality control, scheduling commitments, staffing and budgetary concerns are addressed.

Table 5. Quality Assurance Management Reports

Type of Report	Frequency	Projected Delivery Dates	Person Responsible for Report Preparation	Report Recipients
Integrated Report	Biennial	April	Dave Montali	DEP Leadership EPA General Public
Ecological Assessments	Variable, as watersheds are assessed	Not Applicable	Doug Wood	DEP Leadership EPA General Public
TMDL Reports	Annual	December	Dave Montali	DEP Leadership EPA General Public

## Chapter D Data Validation and Usability

### Section D1 Data Review, Verification, and Validation

Data review and verification is performed in-house to ensure that the data are obtained according to protocol and have been received, recorded, and processed correctly. The data management team (Section C10) works with other Watershed Branch employees to assure that the process is complete.

Data validation is performed by individuals having a less intimate relationship with the data, such as 303(d) list developers and the TMDL modelers. Data are reviewed to determine whether or not they meet the needs of these end users. If the data are found to be deficient in an area, efforts are taken to correct these errors through revisions of field protocols or data evaluation methods.

## Section D2 Verification and Validation Methods

When any of the four major field components is performed, field personnel work from a pre-determined list of sites. This list is maintained in an Excel spreadsheet, which is subsequently used for verification and data entry purposes.

At the completion of an assignment, field crews submit field forms and chain-of-custody forms to the Field Data Manager. These forms are compared to the original list and changes are documented. Coordinates are added to the electronic list and plotted to verify that samples were taken from the correct sites and to verify coordinate data entry. As water chemistry information is received, it is examined for completeness and quality. Paper versions are collated with their respective field forms and electronic results are merged with the existing files. The electronic version is then merged with WAPBASE and paper files are submitted to the Data Entry Manager for additional processing. The Data Entry Manager verifies the correctness of merged data and is responsible for assuring that all keyed-in data are reviewed by a second individual and that all changes are documented within the database. Paper documents are then filed in the central file room at DEP's headquarters. A check-out system is maintained to track files that have been removed for review.

The STORET Data Manager enters these data into the national EPA database. STORET data entry is spot-checked for correctness by the Field Data Manager.

The WAPBASE Data Manager reviews WAPBASE for inconsistencies, verifies sample locations, maintains supportive databases, and performs data reduction and transformation tasks.

The General Data Manager is responsible for importing WAPBASE data into the Decision Database (DDB). Information in the DDB is then used to populate the Assessment Database (ADB).

Data validation is performed by TMDL modelers; 305(b) report preparers and 303(d) list decision-makers. Anomalies or conflicting data are reviewed and corrections are made, if possible. In some cases conflicting information may be an indication of an insufficient sampling plan. If this is true, field protocols are reviewed and revised to address these issues.

## Section D3 Reconciliation with User Requirements

General watershed assessments, pre-TMDL sampling, and the Ambient Water Quality Network are not probabilistic sampling designs and, as such, cannot be subjected to rigorous statistical analysis. However, these activities do require duplicate sampling, which is used to evaluate the ability of the individuals to produce similar data. Macroinvertebrate data from duplicate samples are subjected to precision estimates, the results of which may be used to re-adjust the categories in the WVSCI.

The Watershed Branch's Probabilistic sampling effort was designed after EPA's R-EMAP Program and the data generated through the probabilistic project can be subjected to a multitude of statistical evaluations similar to those used by EPA. The objective of the Watershed Branch's probabilistic project study is to compare the percentage of stream miles affected by a given parameter (i.e. acid mine drainage or sedimentation) with a 90% confidence level. These types of evaluations can be performed at the watershed, ecoregion, or statewide level. Tables and charts will be used to illustrate trends, relationships and anomalies. If information obtained through probabilistic sampling fails to address a specific question, field protocols can be redesigned to incorporate new parameters.

## Appendix A

### Data Quality Objectives Process

**DEPARTMENT OF WATER AND WASTE MANAGEMENT**  
**DIVISION OF WATER AND WASTE MANAGEMENT**

**Data Quality Objectives Process**

**for**

**WATERSHED BRANCH MONITORING ACTIVITIES**



west virginia department of environmental protection

## Data Quality Objective Process

### Step 1. Identify the Problem

The West Virginia Department of Environmental Protection (DEP) established its Watershed Assessment Section in 1996. This section, currently known as the Watershed Branch, consists of managers, engineers, geologists, and biologists. The experience these individuals bring into the program includes laboratory analysis, taxonomy, quality assurance, statistical analysis, and field research. The skills of every individual are considered and utilized during the planning and decision-making processes.

The overall goal of the Watershed Branch is to monitor and assess streams, rivers, and lakes throughout the state to assure these waterbodies are meeting their designated uses. The basic problem that needs to be addressed can be summarized into the following question:

#### **Is this waterbody meeting its uses?**

The Watershed Branch's goal was to apply this question to as many waterbodies as possible. In 1996 existing data for the states' waters were sporadic and varied in quality and level of detail; very few studies offered current information. Therefore, to achieve its goal the Watershed Branch needed to devise methods to effectively monitor the states aquatic resources in a consistent, efficient, and scientifically valid manner. The efforts of the Watershed Branch may be broken down into these components:

- To collect and interpret chemical, physical and biological data from the state's waterbodies in order to obtain current, accurate information on these ecosystems;
- To provide direction to the water quality control efforts of the DEP and other state and federal agencies
- To measure the effectiveness of these agencies' efforts to manage and protect the water resources of the state
- To support stakeholders in the implementation of management and control measures in priority watersheds.

The data collected by the Watershed Branch will be used to identify and assess impaired streams for TMDL development, to identify streams for 303(d) listing or de-listing, to identify and protect high quality streams, and ultimately, to



determine the effectiveness of water management and control efforts. To achieve these goals, the Watershed Branch must provide consistent, comparable data over an extended period of time and develop assessment tools to evaluate complex biological data without insertion of personal bias.

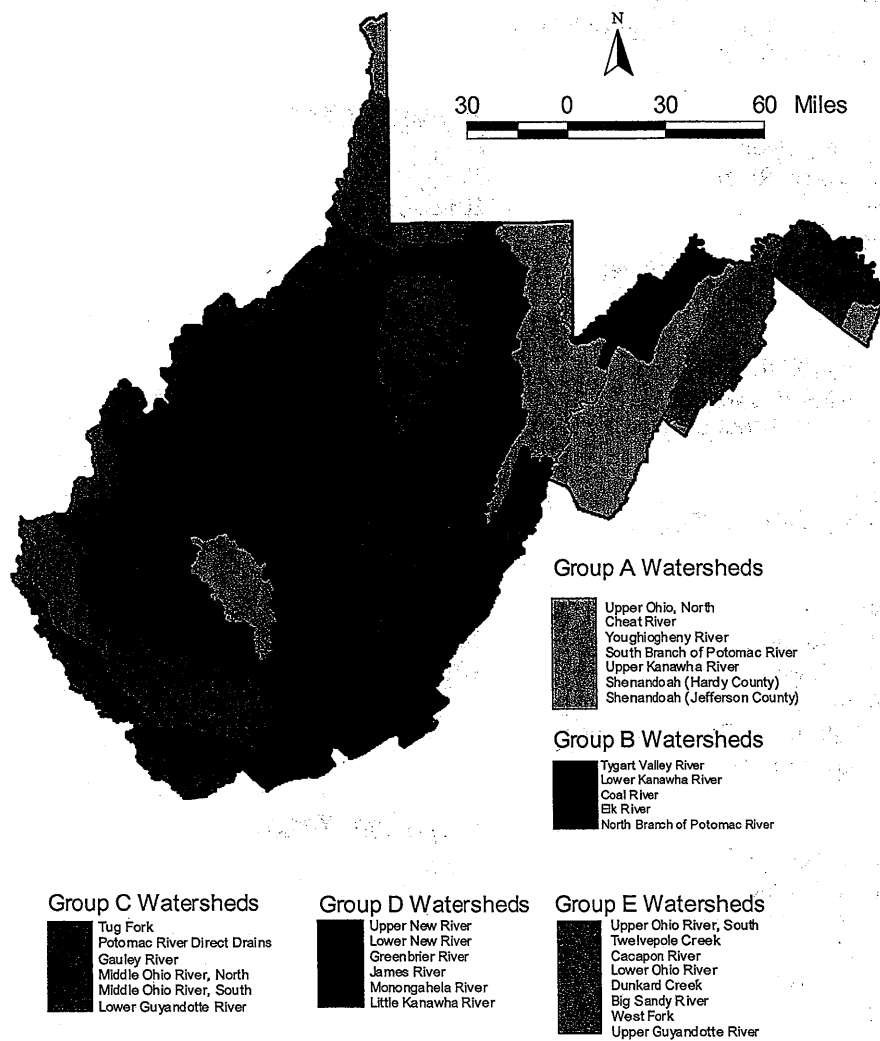
West Virginia has been divided in 32 waterbodies to create manageable work units. These watershed divisions were created using the United States Geologic Survey's eight-digit cataloging unit method. The cataloging units are as follows:

Code #	Watershed Name	HUC Code
1	South Branch of Potomac River	02070001
2	North Branch of Potomac River	02070002
3	Cacapon River	02070003
4	Potomac River Direct Drains	02070004
5	Shenandoah River (Hardy County)	02070006
6	Shenandoah River (Jefferson Co.)	02070007
7	James River	02080201
8	Tygart Valley River	05020001
9	West Fork	05020002
10	Monongahela River Direct Drains	05020003
11	Cheat River	05020004
12	Dunkard Creek	05020005
13	Youghiogheny River	05020006
14	Upper Ohio River, North	05030101
15	Upper Ohio River, South	05030106
16	Middle Ohio River, North	05030201
17	Middle Ohio River, South	05030202
18	Little Kanawha River	05030203
19	Upper New River	05050002
20	Greenbrier River	05050003
21	Lower New River	05050004
22	Gauley River	05050005
23	Upper Kanawha River	05050006
24	Elk River	05050007
25	Lower Kanawha River	05050008
26	Coal River	05050009
27	Upper Guyandotte River	05070101
28	Lower Guyandotte River	05070102
29	Tug Fork	05070201
30	Big Sandy River	05070204
31	Lower Ohio River	05090101
32	Twelvepole Creek	05090102

Each year, the Watershed Branch's data collection efforts will focus on four to eight of these watersheds. Upon the completion of a five-year cycle, each watershed will have been studied and the cycle is then repeated. The watershed sequence is presented below and is illustrated in Figure 1.

<b>Group A:</b>	<b>Assessment Years</b>
Upper Ohio River, North	2001
Cheat River	2006
Youghiogheny River	2011
South Branch of Potomac River	
Shenandoah River (Hardy County)	
Shenandoah River (Jefferson County)	
Upper Kanawha River	
<b>Group B:</b>	<b>Assessment Years</b>
Tygart Valley River	2002
Lower Kanawha River	2007
Elk River	2012
Coal River	
North Branch of Potomac River	
<b>Group C:</b>	<b>Assessment Years</b>
Middle Ohio River, North	2003
Middle Ohio River, South	2008
Potomac River Direct Drains	2013
Tug Fork	
Gauley River	
Lower Guyandotte River	
<b>Group D:</b>	<b>Assessment Years</b>
Lower New River	2004
Upper New River	2009
Greenbrier River	2014
James River	
Monongahela River Direct Drains	
Little Kanawha River	
<b>Group E:</b>	<b>Assessment Years</b>
Upper Guyandotte River	2005
Twelvepole Creek	2010
Upper Ohio River, South	2015
Cacapon River	
Lower Ohio River	
Dunkard Creek	
Big Sandy River	
West Fork	

Figure 1. Watershed Groupings for Assessment within the Five-Year Cycle.



Budgetary constraints place limits on the amount of work that can be accomplished within a given year. The Watershed Branch seeks funding from state and federal sources annually. State budgets are first assembled in August, evaluated by the legislature each spring, and made available to the program in July. Section 106 federal monies are applied for by July and received in October. All Watershed Branch activities are carefully monitored to operate efficiently and within budgetary constraints. A crew of 14 scientists is dedicated to performing the field work. This team is further supplemented during the summer months via temporary employment of college students. Contractual work, such as chemical analyses, organism identification, and TMDL modeling are subjected to a qualifying and bidding process.

## Step 2. Identify the Decision

The principal study question for the Watershed Branch is:

### **Is this waterbody meeting its uses?**

When this question is answered, the following actions may be taken. Depending on the severity of impairment, one or more of these options may apply.

- ▣ Notify Environmental Enforcement or other regulatory entity for immediate resolution. This option is chosen if active violations of state regulations are observed by field crews.
- ▣ Add the waterbody to the 303(d) list of impaired streams
- ▣ Schedule TMDL development
- ▣ Capture assessment data for future action and/or reporting (e.g. Category 1-meeting all uses)

The purpose of the decision statement is to determine if a waterbody is supporting its designated uses. If the waterbody is non-supporting and if the cause of impairment is active, acute, and in violation of state laws, DEP's Environmental Enforcement is notified. If the waterbody is impaired, but the source of impairment is chronic, it is added to the 303(d) list and prioritized according to the established scheduling methodology.

If the waterbody is supporting its designated uses and is considered to be high-quality, it will be protected at a Tier 2 or higher level of detection.

### Step 3. Identify the Inputs to the Decision

In order to address the decision statement, the Watershed Branch requires substantial amounts of chemical, physical, and biological information from a large number of waterbodies. These data need to be obtained in a consistent manner so that comparisons could be made. Existing data were spatially sporadic, outdated, and many were obtained with varying methods; therefore, a fresh dataset was required.

An intense assessment protocol has been developed for use at each site. These assessments include documentation of sampling location, instream and riparian habitat evaluations, benthic macroinvertebrate collections, and the collection of water samples for chemical analysis. Benthic data and habitat evaluations are performed in accordance with EPA's Rapid Bioassessment Protocols (RBP)<sup>1</sup>. Additional field observations were added to the RBP protocols to aid in the decision-making process. Water chemistry is either measured on-site using calibrated instruments or samples are preserved and analyzed by state-certified laboratories in accordance with 40 CFR 136 or SW-846. All information generated by the Watershed Branch is maintained in databases, which are used to expedite decision-making.

The initial round of field assessments (from 1996 through 2000), which focused on wadeable streams, adequately addressed the issues identified by the stakeholders; however, as these data were analyzed, further questions arose. As a result, the Watershed Branch reviews and improves its assessment protocols annually. Examples of such changes include expansions to the on-site observations, new habitat measurement techniques, new aquatic communities, and the inclusion of additional waterbody types (non-wadeable streams, lakes).

Action Levels are values that provide the basis for choosing among the alternative actions (i.e., degree of impairment). Action Levels for water analyses are embodied in the state's water quality criteria. The West Virginia Stream Conditions Index, or WVSCI, is a tool designed specifically for the state of West Virginia to evaluate macroinvertebrate assemblages. The WVSCI was developed using macroinvertebrate data identified to the family level. The Watershed Branch is currently developing a new tool, the Genus Level Index of Most Probable Stream Status (GLIMPSS), to further enhance the interpretation of benthic data. Similar indices will be developed for fish and periphyton communities.

All field personnel must adhere to a set of standard operating procedures (SOPs) to reduce variability and bias during site assessments. These SOPs cover

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<sup>1</sup> Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers: Periphyton, Benthic Macroinvertebrates and Fish. Second Edition. 1999. U.S. Environmental Protection Agency. EPA 841-B-99-002.

every aspect of on-site assessment including equipment calibration and maintenance, sample collection and preservation, and guidelines for completing habitat assessment forms. Off-site chemical analyses are performed by state-certified laboratories and are handled and analyzed in compliance with 40 CFR 136 and SW-846.

#### Step 4. Define the Boundaries of the Study

The target population includes any or all of the states water resources. Spatial boundaries for most studies are defined in the five-year watershed cycle presented in earlier sections.

Pre-TMDL development samples are collected each month from July through June. This procedure assures that data will be obtained during a variety of environmental conditions. While the actual sampling dates are not randomly determined, care is taken to collect information during high, low, and normal flow conditions.

Probabilistic and Watershed Assessments are performed seasonally. Samples for these projects are collected only when conditions are favorable for effective collection of the target biota. Benthic macroinvertebrates are sampled from April through October. New research indicates that benthic sampling may begin earlier in the season, but should be avoided in June due to the high numbers of emerging adults.<sup>2</sup> Fish populations in wadeable streams may be collected year round; but larger stream sampling is only effective during the warmer months when fish are more active. Regardless of stream size, attention must be paid to the applicable index period. Data collection for both projects is suspended during periods of high turbidity and/or high flow as these conditions can obscure important observations and may create unsafe conditions.

The Ambient Water Quality Monitoring Network, which emphasizes larger streams and rivers, does not comply with the watershed cycle. All samples in this network have been sampled quarterly; and, beginning in 2006, these sites will be visited 6 times per year (bi-monthly).

Data are reviewed periodically to assure the results are providing the information required by the specific projects. Pre-TMDL development sampling networks are examined mid-cycle (January). Sites may be added to or dropped from the network at this point. Probabilistic sampling is evaluated annually

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<sup>2</sup> It should be noted that an expansion of the sampling time frame for benthic macroinvertebrates would require a re-evaluation of the WVSCI to determine its effectiveness in evaluating "off-season" data.

and field protocols are adjusted as deemed necessary. Watershed Assessment samples are critically evaluated in conjunction with 303(d) list preparation.

## Step 5. Develop a Decision Rule

Data collected for the Watershed Assessment and Probabilistic programs are one-time events, therefore any initial decisions made from this information assumes that the results are the "true" value. Two parameters are currently used in the decision-making process: water quality and benthic macroinvertebrate populations. Analytical values for water are examined for violation of the state's water quality criteria. Benthic macroinvertebrate data are examined via the WVSCI score. Several decision scenarios may arise during this process. These scenarios and the theoretical decisions associated with them are presented below. Older data may also be considered at this point, as they may be used to determine the frequency or persistence of violations.

1. **Water quality is unimpaired, benthos are unimpaired (WVSCI >68)** - Consider waterbody for placement in Category 1 and also consider site as a potential reference.
2. **Water quality is unimpaired, benthos are of uncertain status (WVSCI between 68 and 60.6)** - Examine assessment data in more detail before making a decision. The observations documented by the field crews can often reveal conditions - both natural and human-induced - that could depress benthic populations without causing violations of water quality criteria.
3. **Water quality is unimpaired, benthos are impaired (WVSCI <60.6)** - Include waterbody on 303(d) list, establish priority for TMDL development.
4. **Water quality is impaired, benthos are impaired (WVSCI <60.6)** - Include waterbody on 303(d) list and establish priority for TMDL development.

It is acknowledged that the addition of new parameters, such as fish and periphyton assemblages, will further complicate this process.

The focus of pre-TMDL development is on streams that have already been subjected to a decision-making process. Unimpaired or unassessed streams within the study are may also be included to provide additional information for TMDL development. At the conclusion of the Pre-TMDL sampling process, these data are submitted to contracted TMDL modelers for further analysis. The final TMDL will determine what action is to be taken.

The Ambient Water Quality Monitoring Network is used to evaluate temporal and/or spatial long-term trends. These data are subject to periodic review and evaluation. When violations are observed the waterbody is considered for 303(d) listing.

## Step 6. Specify Tolerable Limits on Decision Errors

Every research effort has an element of uncertainty. Natural variability and limitations of measuring instruments will prevent the collection of “true values” for a given set of parameters. The selection of an appropriate sampling design and compliance with specific field and laboratory protocols will help to reduce the degree of error that may occur. The allowable degree of error and the consequences of these errors must be addressed. This effort is accomplished by the development of a baseline hypothesis, a definition of the areas of uncertainty, and tolerable limits for decision errors.

The baseline hypothesis for Watershed Branch projects is:

**Measured water quality values are below state water quality criteria AND the WVSCI score is greater than 68.**

The burden of proof is in *rejecting* this hypothesis; that is, it must be proven that a waterbody has violations and/or has a low WVSCI score. For the needs of the Watershed Branch, a relatively small amount of data can result in rejection of this hypothesis.

Two types of decision errors can result from this hypothesis. These errors are most likely to occur when study results approach the Action Levels: The closer the data are to water quality criteria or to the WVSCI score of 68, the more likely it would be to take improper action.

The first kind of error is a *false acceptance* of the baseline (also known as a Type II error). This type of error occurs when one assumes the baseline is true, when in reality it is false. False acceptance occurs when a waterbody is determined to be unimpaired, when in actuality it *is* impaired. The consequences of making a false acceptance error would be to take no action on a waterbody that needed to be subjected to pollution abatement/control procedures. If a false acceptance occurs, a stream that is only slightly impaired would not be subjected to further investigation.

The second type of error is a *false rejection* of the baseline (also known as a Type I error). False rejection errors occur when one assumes the baseline is false when it is actually true. A false rejection occurs when a waterbody is flagged as impaired, when it is actually unimpaired. If a false rejection occurs,

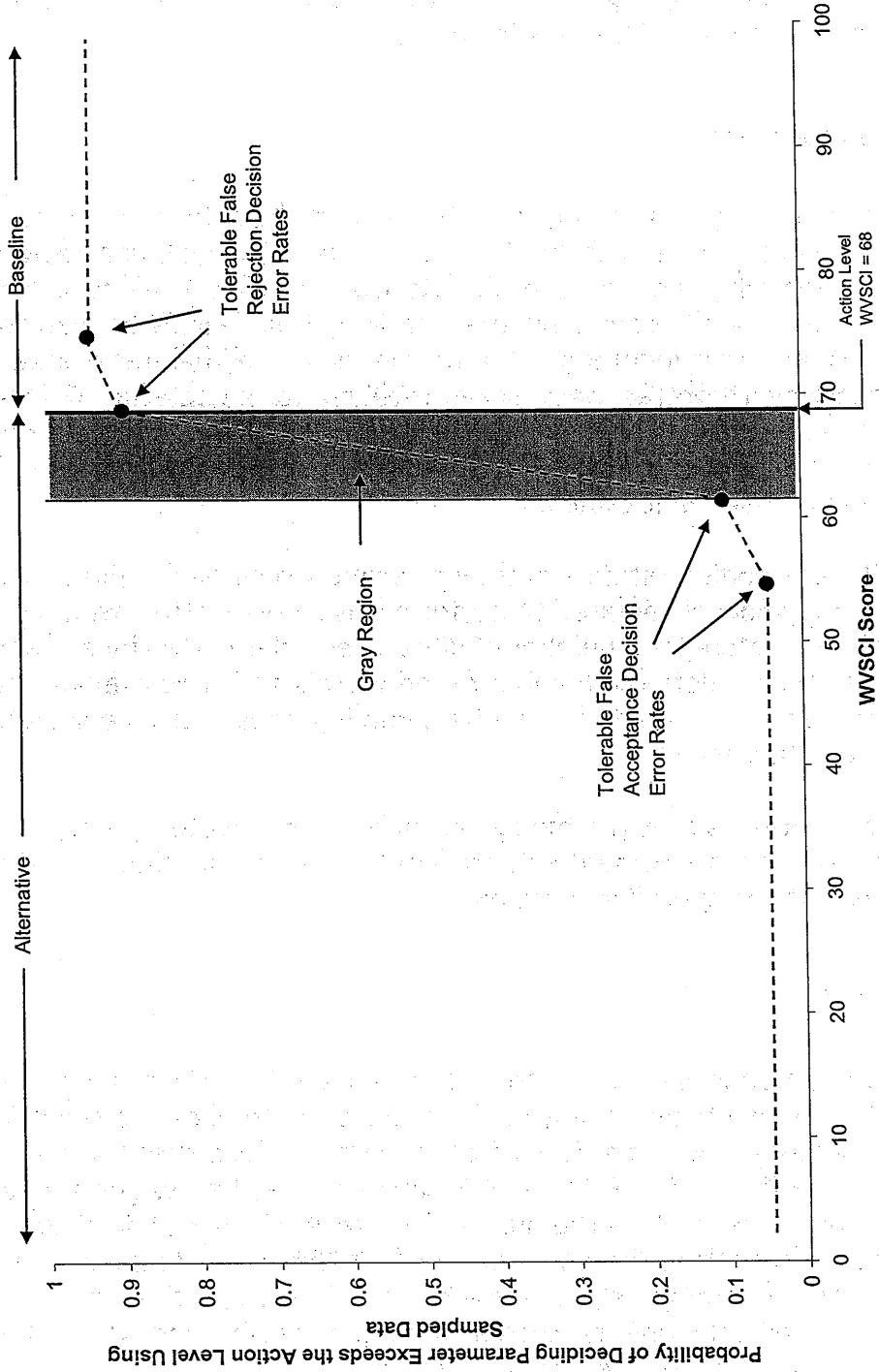


an unimpaired stream would be subjected to additional sampling, which will provide evidence that an error had been made.

In addition to error possibilities discussed in the preceding paragraphs, there is a region known as the “gray area” where a decision is too close to call. As a measured water quality value approaches the state criterion (or the WVSCI score approaches 68) the probability of making an incorrect decision is more likely. The “gray area” is the region where costs of further sampling and analysis outweigh the consequences of making an incorrect decision. Since these errors happen as the data approach the action level, it is likely that these minimally impaired waterbodies would be identified as “low priority” and not subjected to immediate TMDL development or other actions.

A Decision Performance Goal Diagram for the WVSCI is presented in Figure 2. This diagram is a graphical depiction of the baseline condition, gray area, and the tolerable limits on decision errors. Decisions made using water quality criteria are more complex and cannot be easily graphed. However, a Decision Performance Goal Diagram for water quality would be similar to the diagram for the WVSCI.

Figure 2. Performance Goal Diagram for the WWSCI.



## Step 7. Optimize the Design for Obtaining Data.

The Watershed Branch has developed four projects to meet the various needs of the agency. The sampling design and key assumptions supporting these designs are presented in the following paragraphs.

### Watershed Assessments

The goal of watershed assessment is to obtain current data throughout a given watershed, in accordance with the five-year cycle. The sampling design was developed to obtain a large amount of data from as many streams as possible. Existing data are used to select sample locations, but unassessed waterbodies are also considered for inclusion. Measurements include habitat evaluations, benthic macroinvertebrates, periphyton, fecal coliform bacteria. Grab samples for other parameters may be taken at the discretion of the field personnel. Sampling frequency is one visit per site per year. A site may or may not be reevaluated in subsequent cycles.

Ideally, all waterbodies within a given watershed would be sampled; however, the costs and personnel required to perform this level of effort would be prohibitive. Therefore, the number of sites selected is driven by a "sampling budget", which considers the number of individuals and time available to assess a watershed. To further expedite sampling, most sites are selected in areas with easy access.

The key assumption is that the biological, habitat, and water quality components of each assessment will present an overall snapshot of the health of the stream at the specified location.

### Probabilistic Sampling

Probabilistic sampling is designed to address some of the shortfalls of general watershed assessments by employing a statistical method of site selection into the sampling design. A model is used to randomly choose sites based on a pre-determined set of criteria. Probabilistic sampling may be designed to address a specific set of issues, such as the number of stream miles impaired by water quality violations or the differences between ecoregions. Measurements are the same as for general watershed assessments, but also include additional habitat evaluation components (such as the riffle stability index). A standard set of water quality parameters is also obtained at each site.

Probabilistic sampling is performed state-wide within a designated time frame. Six samples are collected from each watershed annually. At the completion of the five-year cycle, thirty or more sites will have been sampled from each watershed<sup>3</sup>. Statisticians indicate that thirty samples are sufficient for accurate analysis of environmental data.

The key assumption is that the thirty sites visited will represent the overall conditions of the watershed under investigation.

### Pre-TMDL Sampling

Results of pre-TMDL sampling are applied to models for TMDL development. The accuracy of these models is dependant on the amount of data available. Sampling frequency is once per month for twelve months. This plan allows the Watershed Branch to capture data under a variety of flow regimes and weather conditions. The data collected from each sampling site will vary, depending on the suspected cause of impairment. These data may include mine drainage, acid rain, and/or nutrient loading parameters; biological sampling; fecal coliform bacteria; and flow. Data generated by watershed assessments, probabilistic sampling, and external sources are considered in the site selection process. The sample design may also include pollution sources such as mine discharges.

The key assumption is that twelve sampling events, occurring over the course of a year and under various conditions, will accurately represent the natural conditions of the waterbodies under investigation.

### Ambient Water Quality Monitoring

The Ambient Network was established on the state's larger streams and rivers to identify long-term temporal and/or spatial trends. This project was established in the 1940's and the number of stations and sampling frequency have gone through numerous, undocumented changes throughout the years. The current sampling design consists of 25 fixed stations. In recent years, sampling frequency has been quarterly. However, beginning in 2006 these sites will be visited bi-monthly (6 times per year). Sampling protocols include a standard set of water quality analyses, field observations, and flow measurements (direct measurement or via gages). Grab water samples are collected at the surface. Annual aquatic organism collections have recently

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<sup>3</sup> Watersheds having fewer than thirty named streams were merged with larger watersheds.

been reinstated for the Ambient Network<sup>4</sup>. Benthic macroinvertebrates are collected from non-wadeable sites using Hester-Dendy multiplate samplers. Sites having wadeable, riffle habitat are subjected to standard Watershed Assessment protocols.

The key assumption for the Ambient Network is that the selected sampling frequency and parameters will provide sufficient information for periodic trend assessments.

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<sup>4</sup> Macroinvertebrate samples were collected annually beginning in the 1970's; however, this component was dropped in the late 1980's.

Appendix B

Watershed Branch

Standard Operating Procedures Manuals

**West Virginia Department of Environmental Protection  
Watershed Branch  
Standard Operating Procedures**

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- A. YSI and Hydrolab Calibration
  - B. Instructions for Completing the Habitat Evaluation Form
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- Supplemental Documents
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# SOP A. YSI AND HYDROLAB OPERATION & CALIBRATION

## **INTRODUCTION**

The following procedures are an overview of YSI calibration for a YSI 600XL Sonde/650MDS display combination and Hydrolab Quanta G. Consult the owner's manuals for specifics or information on configurations other than these and for details on maintenance and trouble-shooting. These procedures assume the user has a basic knowledge of the instrument.

These directions are not intended for first-time users. Individuals with no prior experience should calibrate with the assistance of an experienced user.

All calibration adjustments are documented in a permanent logbook. The date and time of calibration, name of the calibrator, the identification number of the unit, battery voltage and all adjustments/maintenance must be documented.

**Note:** Rinsing the probe is a procedure that is frequently performed during calibration. To rinse the probe, install the calibration cup (which is the same as the storage cup on YSI and Quanta G sondes) and add about 1/2 cup of rinse solution, as specified in the directions (usually deionized water). Seal the open end of the calibration cup with the screw cap or rubber lid and shake the probe for 30 seconds. Discard rinse water and repeat according to directions.

All calibrations are performed with the probes in the pointing upward and at temperatures as close to room temperature as possible (25°C).

## **I. YSI 600XL Sonde/650MDS Display Unit Calibration**

These directions are very similar to the older Scout 2 Hydrolab and newer Quanta G directions. However, individuals with no prior experience should calibrate with the assistance of an experienced user.

### **1) YSI Display Unit**

The YSI display unit uses a series of escapable menus in conjunction with several keys in the calibration process. Become familiar with the enter (which looks like a left arrow), escape, scroll, and alpha-numeric keys as these will be the most often used.



### *Maintenance of YSI Display Unit*

The YSI display unit runs on a 4 alkaline C-cell battery system contained within the display unit. The battery power left is displayed on the screen.

Also of importance is the fact that the results of calibration for YSI units are stored in the sonde itself, not in the display unit. Switching the sonde and display units will not effect calibration. This may be especially helpful as one can calibrate several sondes with only one display unit as others may be recharging.

The display unit also features a Date/Time and an auto-shutoff function, which may be modified by selecting "System Setup" in the main menu and then selecting the appropriate function to modify.

## **2) Dissolved Oxygen**

### **A) Probe Calibration**

1. Remove the threaded lid to the calibration cup. Unlike the Hydrolabs, it is not necessary to dry the membrane on the D.O. probe by blotting it with a soft cloth or tissue, but rather only make sure that the membrane is **not inundated with water**. Also, check the membrane for wrinkles, tears, bubbles, dirt, etc. and replace membrane, if necessary.
2. Reattach the calibration cup to the sonde and add no more that 1/8-inch of DI water. Try not pour water on the membrane, but if it does get wet, just make sure that the membrane is not totally inundated with water. Make certain that the DO and Temperature probes are not immersed in water.
3. Cover the calibration cup with the lid and engage only 1 or 2 threads.
4. Let the unit sit for about 10 minutes so that the air inside the cup will saturate with water.
5. Turn on the unit and use the **Up** or **Down** keys to scroll to "Sonde Menu" and press **Enter**. Select "Calibrate" and press **Enter**.
6. Scroll to select "Dissolved Oxy" and press **Enter**.
7. Select "DO %", press **Enter**. One must keep in mind that this is actually calibrating based on O<sub>2</sub> air saturation, not water saturation.

8. Type in the Barometric Pressure displayed by the unit in the bottom right of the screen using the alpha-numeric pad; press **Enter**. Wait for all readings to stabilize; this may take up to 40 seconds. Record the initial or pre-calibration DO, temperature, and % air saturation in logbook.
  
9. Press **Enter** to finish calibration. Record the final or calibrated DO and % air saturation in log book. The final % air saturation should be within the range of 98% air saturation at the lowest WV elevations to 83% at the highest WV elevations (+/- 2 %). The probe should typically not read above 100% air saturation as this only occurs at sea level. A 100% reading may also be caused at low WV elevations by a high-pressure front or unusual weather in the area. Consult the attached sheet for air saturation values that should be found at different elevations or Appendix D (page 227) from the YSI operating manual (Figure A-1). YSI probes may be calibrated at lower elevations and then brought to a higher elevations and still be accurate. However, calibration at an extreme elevation and transport to a lower elevation may require a recalibration at the lower elevation. If the barometer reading is extremely unusual for your local elevation, the internal barometer may require recalibration in the lab by a person familiar with that procedure.

**Figure A-1. From Appendix D Table 2 of the YSI operating manual (page 227)**

Pressure (mm Hg)	Altitude (ft)	Expected % Saturation (+/- 2%)
760	0	100
752	278	99
745	558	98
737	841	97
730	1126	96
722	1413	95
714	1703	94
707	1995	93
699	2290	92
692	2587	91
684	2887	90
676	3190	89
669	3496	88
661	3804	87
654	4115	86
646	4430	85
638	4747	84
631	5067	83

Elevation at Harpers Ferry 249 ft and at Spruce Knob 4862 ft.

10. Press **Enter**. And it will take you back to the DO Calibration Menu.

## B) Probe Maintenance

The membrane on the DO probe should be examined for fouling and bubbles before calibration and during use. If the membrane is torn, dirty or wrinkled, or if there are bubbles under the membrane, the membrane must be replaced. YSI recommends the membrane be replaced at least every 30 days. **The membrane should be replaced 24 hours before use or calibration. In an emergency, a minimum of 30 minutes must elapse prior to use.**

To replace the membrane, remove the O-ring and old membrane and shake the remaining electrolyte (KCl solution) out of the probe. Add a few drops of fresh KCl solution to the probe. The tip of the probe should be filled to create a positive meniscus (an "outie"), and should be free of bubbles. Hold new membrane between thumb and probe body. Use your free hand to stretch the membrane up, over, and down the opposite side of the probe. Secure the loose end with your forefinger. Roll the O-ring over the tip of the probe without touching the membrane with your finger. Cut off excess membrane. Document any membrane replacement in the logbook.

## C) Probe Diagnostic

To check the quality of the calibration or diagnose a potential problem with the DO probe, an advanced function called DO charge may be used.

1. Press **Esc** to get the Main Menu.
2. Use the **Up** or **Down** keys to scroll and select "Report".
3. Scroll down and select "Dochrg" and press **Enter**. When this is done, the symbol to the left of "Dochrg" should change from an empty to a black circle.
4. Press **Esc** twice to get the 650 Main Menu. Scroll up to "Sonde Run" and press Enter.
5. A new parameter should be visible on the screen called "DOc". If the probe is in adequate condition and calibrated successfully, the number should range from 25 - 75 with a score of 50 being optimum.
6. If the probe reads in this range, then simply repeat this procedure to turn off the Do charge function (the black circle will change back into an empty circle).

If the DO charge is in the low end of the range or below this range, the KCl solution under the membrane may be contaminated with water. In this case the membrane and solution should be replaced.

If the DO charge is in the high end of the range several things may be wrong. First, the highly malleable Au electrode may be distorted or the silver-plating on the electrode may be "tarnished" and gray looking. In this case, the electrode may be reconditioned by buffing it using one of the **YSI provided buffing discs only**. THIS SHOULD ALSO BE DONE ONLY WITH STRICT ADHERENCE TO THE DIRECTIONS PROVIDED IN THE MANUAL FOR USING THIS BUFFING DISC ON THE PROBE SURFACE. IT MAY BE NECESSARY TO CONSULT WITH AN YSI REPRESENTATIVE BEFORE ATTEMPTING THIS ACTION. A second possible cause of a high DO charge reading are cracks around the electrodes as a result of drying and rewetting of the surface. If this is the case, then the DO probe may need to be replaced.

**The DO probe accuracy range is +/- 0.2 mg/L (or ppm) O<sub>2</sub> or 2% of the reading (whichever is greater). The range for % saturation is or +/- 2 % or the reading or Air Saturation (whichever is greater).**

### 3) Conductivity

#### A) Probe Calibration

1. Remove the lid on the calibration cup and use the special brush designed to fit inside the conductivity probe's 2 end ports, vigorously scrub each port 5-10 times.
2. Rinse the probe 3 times with deionized water.
3. Rinse the probe 2 times with a small amount of conductivity standard in the **1000-5000** microSiemens range. (The exact concentration of the standard will be written on the bottle.).
4. Fill cup with conductivity standard to within a centimeter of the top of the cup. Make sure that there are no bubbles in the measurement cell of the specific conductance sensor. Record the concentration of standard used to calibrate in the logbook.
5. Press **Escape**. Scroll to **conductivity**; press **Enter**.

6. Scroll to **SpCond**; press **Enter**.
7. Type in concentration of standard in **MILLI**Siemens (not MicroSiemens. 5000 microSiemens = 5.000 milliSiemens. Press **Enter**.
8. Record the initial or pre-calibration readout in logbook. Press **Enter** to calibrate and record the final or calibrated readout in logbook. Press **Enter** again to continue back to the conductivity menu.

#### B) Probe Maintenance

The openings that allow fluids to access the conductivity electrodes should be cleaned regularly (once a month at most) using the small acrylic brush included in the YSI calibration kit. Dip the brush in clean water and insert it into each hole 20-30 times. A mild detergent may be used with the brush, if deposits have formed on the electrodes.

#### C) Probe Diagnostic

The conductivity probe on an YSI sonde can be checked using a function called Cal Constants.

1. Press **Esc** to get the Main Menu.
2. Scroll down and select "Advanced" and press **Enter**.
3. Scroll down and select "Cal Constants" and press **Enter**. The reading next to the "Cond:" should range from 4.5 – 5.5. **IF THE READING IS NOT WITHIN THIS RANGE CONSULT THE YSI OPERATION MANUAL OR AN YSI REPRESENTATIVE.**
4. To escape from this screen, press Esc repeatedly until the Main Menu appears.

**The Conductivity probe accuracy range is +/- 0.5% +/- 1 uS/cm . For example, a solution that is 1000 MicroSiemens, the range would be 1000 x 0.005 +/- 1 MicroSieman or 5 +/- 1 MicroSieman.**

#### 4) pH

##### A) Probe Calibration (a three-point style calibration)

1. Press **Escape** to get to Calibration mode.
2. Rinse probe three times with DI water.
3. Scroll down to **ISE1 pH**; press **Enter**.
4. Scroll down to **3 Point**; press **Enter**.
5. Rinse probe twice with DI water and once with 7.0 buffer solution.
6. Fill calibration cup with 7.0 buffer solution to within a centimeter of the top of the cup.
7. Type in 7.00; press **Enter**. Allow readout to stabilize.
8. Record the initial or pre-calibration readout. Press **Enter** to calibrate.
9. Record the final or calibration readout and press **Enter** again.
10. Rinse probe 2 times with deionized water and once with pH 4.00 buffer solution.
11. Fill calibration cup with 4.00 buffer solution to within a centimeter of the top of the cup.
12. Type in 4.00; press **Enter**. Allow readout to stabilize.
13. Record the initial or pre-calibration readout; press **Enter** to calibrate.
14. Record the final or calibration readout and press **Enter** again.
15. Rinse probe 2 times with deionized water and once with pH 10.00 buffer solution.
16. Fill calibration cup with 10.00 buffer solution to within a centimeter of the top of the cup.
17. Type in 10.00; press **Enter**. Allow reading to stabilize.

18. Record the initial or pre-calibration readout; press **Enter** to calibrate. Record the final or calibrated readout. Press enter to return to the pH calibration menu.
  
19. Add a *very* small amount of tap water (just enough to keep the air inside the cup moist) to the storage cup. **THE STORAGE WATER SHOULD NOT COVER THE PH OR DO PROBE WHEN THE SONDE IS HORIZONTAL.**

## B) Probe Maintenance and Troubleshooting

Sometimes slow response times or instability with the values (jumping as much as +/- 1.0 unit during calibration or field measurements) are observed with the pH readings. This may be caused by a number of factors and may or may not be indicative of a bad probe.

One consideration is the age of the probe. If a probe is 18 months or older, then it has reached its life expectancy and more than likely, it is dead. To check the probe's manufacture date, look at the first three characters of the alpha numerical stamp on the side of the probe. The first two digits represent the year and the third character the month (e.g. 02G is July of 2002 as G is the seventh letter of the alphabet).

Another factor to consider is the temperature probe. The calculation of the pH by the sonde is a temperature dependent calculation. If the temperature probe is malfunctioning, then it may appear as if the pH probe isn't working right. Be sure to check the temperature to see if it returning a reasonable value. If it is not, then the temperature/conductivity probe may need to be replaced.

Water or sealant grease can also get in the connector when replacing a probe and can cause malfunctions and erratic readings. When replacing a pH probe, dry off the probe and sonde as much as you can before removing the probe to make sure water doesn't enter the fitting. Also, remove the pH probe with the sonde upside down so that water can not run into the connections. Once removed, look inside the connector end of the probe and sonde to see if there is any water or grease in the fitting. If so, remove it with a can of compressed air and/or a paper towel. The important thing is to dry it out as much as possible. If there is excessive grease, then try to remove it with a towel. If the grease cannot be removed, an YSI maintenance expert may need to use a solvent for to break up the grease. Once dry, reconnect the probe using very little grease around the upper o-ring near the threads. A very thin coat making the o-ring look wet is sufficient for a proper seal.

Cleaning is required when response becomes slow or when deposits build up on the surfaces. To clean the glass bulb, remove the probe and use a soft cloth or tissue to wipe foreign material from the glass bulb and platinum button. Then use a moisten cotton swab to GENTLY remove any material blocking the reference electrode junction. DO NOT WEDGE THE SWAB TIP BETWEEN THE GUARD AND THE GLASS SENSOR.

If response is still slow, soak the probe 10-15 minutes in clean water containing dishwashing liquid. Then wipe the probes gently with a cotton swab moistened with the cleaning liquid. Rinse in clean water, wipe once more with a clean swab and rinse again.

If response times continues to be slow, the probe may be cleaned in a 1:1 chlorine bleach solution for 1 hour. YSI recommends this procedure ever 6 – 12 months if the probe does not work well. This is usually as result of extreme conditions in which fowling of the probe is more probable.

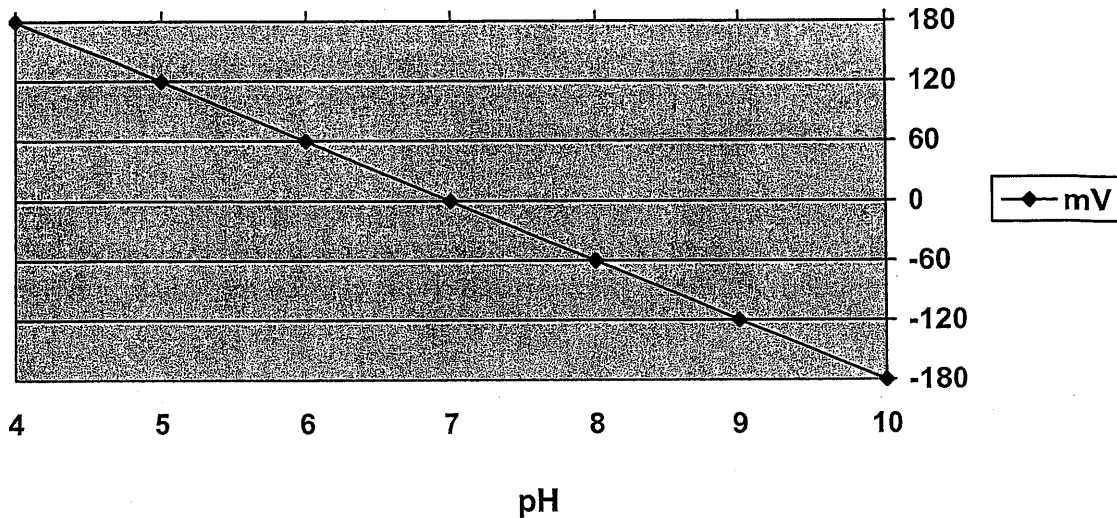
Finally, if the probe still does not respond well, the may be soaked in one molar HCl for 30-60 minutes. THIS SHOULD BE USED AS A LAST RESORT METHOD ONLY. REFER TO THE YSI OPERATING AND MAINTENANCE MANUAL FOR DETAILS ON THESE PROCEDURES OR CONSULT AN YSI REPRESENTATIVE.

#### C) Probe Diagnostic (Nernst Equation Calculation)

The pH probe on an YSI sonde operates using the Nernst Equation (see Figure A-2 below). Simply put, a line running from 4 to 7 (or 7 to 10) pH on the x-axis should increase 180 mV (or 60 mV/pH unit) from 7 to 4 or (decrease 180 mV from 7 to 10) pH on the y-axis. See illustration below. If this slope flattens, the pH probe will lose resolution. This slope is a result of the probe condition as well as the quality of the calibration. A function called pH mV may be used to check this slope.



Figure A-2. The Nernst Equation



The Nernst Equation may be calculated by following these steps:

1. Press **Esc** to get the Main Menu.
2. Use the **Up** or **Down** keys to scroll to Report and press Enter.
3. Scroll down and select "pH mV" and press **Enter**. When this is done, the symbol to the left of "pH mV" should change from an empty to a black circle.
4. Press **Esc** twice to get the 650 Main Menu. Scroll up to "Sonde Run" and press Enter.
5. A new, second parameter called "pH mV" should be visible on the screen that reads well beyond 14 and may be positive or negative.
6. Use the rinse procedures from the pH calibration above to introduce 7.00 buffer solution to the probe. Write down the second "pH" reading for the 7.00 buffer solution. It should be between -40 and 40, but may be slightly more (-50 to 50). The reading should stabilize within the aforementioned values in less than 30 seconds.
7. Repeat the rinse procedures for either 4.00 or 10.00 buffer solution. For either solution, the new reading should have a difference of 180 from the initial 7.00 buffer solution reading.

For example, if an initial reading at 7.00 was -20.0, then the second reading at 4.00 should be around 160 (+/- 40) or, if using 10.00, the second reading should be around -200 (+/- 40).

8. If the probe reads in this range within 30 seconds, then the probe is ok and simply repeat this procedure to turn off the pH mV function (the black circle will change back into a –). If the probe reads outside this range or takes longer than 30 seconds, two things may be wrong. First, the calibration may be off and the calibration procedure should be repeated to check for this. Unfortunately, if recalibration does not correct the problem, this is an indication that the KCl solution inside the probe is contaminated with water and the whole probe will need to be replaced. *See Sonde Storage below for prevention of contamination of the KCl solution inside the pH probe.*

**The pH probe accuracy range is +/- 0.2 pH units (i.e., 6.8-7.2 is an acceptable reading for 7.0 buffer solution.**

#### 5) Temperature

**The Temperature probe accuracy range is +/- 0.15<sup>0</sup> C.**

#### ***Tips for usage of YSI probes in the field:***

- When deploying a probe into the water, give it a little tap or shake once submerged. This will help jar loose any air bubbles inside the conductivity probe that will bias a reading.
- If taking readings from a water sample in a container (e.g., a bucket), make sure to keep the hole on the conductivity probe away from the edges of the container as this may cause stray signals from the probe and result in an inaccurate reading.
- Make sure that all probes are submerged adequately.
- If a DO probe is suspected of being out of calibration, check the DO charge reading as well as the % air saturation. If the air saturation is not within an expected range for your current elevation, recalibration at that elevation may be necessary. It is also possible that the internal barometer needs recalibration.
- When storing the sonde between sites or sampling events, only a few drops of water inside the cup is necessary to keep the air (and membranes) inside the cup moist. **DO NOT STORE THE SONDE WITH A FULL CUP OF WATER, AS THIS WILL LESSEN THE LIFE OF THE pH PROBE.**

- Always make sure sand and other particles are kept clear of the threads on the sampling weight, cap, storage cup, and sonde itself. These threads are plastic and will strip if sand is caught in the treads while screwing these parts on and off.

### ***YSI Sonde Storage***

The pH probe on an YSI sonde operates using a polypropylene wick from the water (or sampling) side to a concentrated KCl side sealed inside the probe. To increase the life of a probe, proper storage of the probe must be implemented.

If the probe is being stored for a **short period of time**, place only a minute amount of water (1/8<sup>th</sup> of an inch is probably too much) in the cup for storage making sure that no water will inundate the pH probe. The other probes (e.g., the DO probe) require only moist air to maintain proper function. A small damp sponge inside the cup would be adequate for such storage.

For **long-term storage** (e.g., over winter), it is recommended that the cup be filled with a concentrated KCl solution. This will lengthen the life of the probe and help maintain the concentration of KCl inside of the pH probe. **TO OBTAIN THE CONCENTRATION OF KCL SOLUTION, CONSULT THE YSI OPERATING MANUAL OR AN YSI REPRESENTATIVE.**

## **II. Hydrolab Quanta G Calibration**

These directions are very similar to the older Scout 2 Hydrolab directions. However, individuals with no prior experience should calibrate with the assistance of an experienced user.

### **1) Quanta G Display Unit**

The Quanta G display unit uses a series of escapable menus in conjunction with several keys in the calibration process. Become familiar with the enter (which looks like a left arrow with a right angle), escape (Esc ∞), on/off (O | I), and arrow keys as these will be the most often used.

#### *Maintenance of Quanta G Display*

The Quanta G runs on 3 C batteries. Replace the C batteries as required. The Quanta G System provides at least 15 hours of continuous operation on one set of new batteries. A Battery Low icon will show the battery status

## 2) Dissolved Oxygen

### A) Probe Calibration

1. Remove calibration cup from probe and dry the membrane by blotting with a soft cloth or tissue. Check the membrane for wrinkles, tears, bubbles, dirt, etc. and replace membrane, if necessary.
2. Attach calibration cup to the Quanta and fill cup with room temperature tap water until the water surface is just level with O-ring on the D.O. probe. Do not pour water on the membrane. If the membrane gets wet, blot dry with a soft cloth or tissue.
3. Cover the calibration cup loosely using the black calibration cup cover placed upside down on the calibration cup.
4. Let the unit sit for about 10 minutes so that the air inside the cup will saturate with water.
5. Turn on the Quanta G using the **O | I** key and allow the D.O. reading to stabilize. If the circulator is on, press the **Esc** ∞ key to toggle the circulator off so that it doesn't splash the water in the cup onto the membrane. Record the initial or pre-calibratino readings (mg/L) into the logbook. Also record the initial readout for temperature.
6. Press the **enter** key to toggle to the next screen and record the initial or pre-calibration % DO saturation in the logbook.
7. After power-up the Display's "Screen" icon in the lower center of the screen is blinking. Press either of the **arrow** keys to cause the "Calib" icon to blink instead of "Screen". Press the **enter** key to select calibration. Use the **arrow** keys to cause "DO" to blink and the press the **enter** key.
8. Determine the barometric pressure for entry as the calibration standard and record in the logbook. *Use the local barometric pressure. Many local weather bureaus correct the barometric pressure to sea level. Consult the operating manual for formulas to convert from sea level barometric pressure to local barometric pressure.*
9. Press the **arrow** keys to raise or lower the barometric pressure to match the calibration standard.

10. Press the **enter** key to finish calibration of Dissolved Oxygen. If the unit rejects the calibration, the display will show "FAIL" before returning to the "Calib" screen.
11. Press the **Esc**  $\infty$  key to return to the real-time data screen. Record the final or post-calibration D.O. readings into the logbook. Press the **enter** key to toggle to the next screen and record the final or post-calibration % DO saturation in the logbook.

## B) Probe Maintenance

If the D.O. will not calibrate, the membrane may be torn, wrinkled, dirty, damaged, or a bubble may be trapped in the probe. The membrane should be replaced whenever these conditions are observed. Frequent replacement of membranes can also lengthen the life of the probe.

To change the membrane, remove the calibration cup. Remove the O-ring that holds the membrane on the probe. Shake out the old electrolyte solution, rinse the probe with electrolyte solution, and refill with fresh electrolyte until a positive meniscus rises above the probe surface. Make sure there are no bubbles in the probe. Install the new membrane (don't stretch the membrane while doing this), and replace the O-ring. If possible allow the probe to soak overnight in tap water to acclimate to its new shape.

**The DO probe accuracy range is +/- 0.2 mg/L (or ppm) O<sub>2</sub> at ≤ 20 mg/L or +/- 0.6 mg/L (or ppm) O<sub>2</sub> at >20 mg/L.**

## 3) Conductivity

### A) Probe Calibration

1. Remove the lid on the calibration cup and rinse the probe 3 times with deionized water.
2. Rinse the probe 2 times with a small amount of conductivity standard in the **1000-5000** microSiemens range. (The exact concentration of the standard will be written on the bottle.).
3. Fill cup with conductivity standard to within a centimeter of the top of the cup. Make sure that there are no bubbles in the measurement cell of the specific conductance sensor. Wait for the readings to stabilize. Record the concentration of calibration standard used and the initial or pre-calibration specific conductance readings in the logbook.

4. Press either of the **arrow** keys to cause the "Calib" icon to blink instead of "Screen". Press the **enter** key to select calibration. Use the **arrow** keys to cause "SpC" to blink and then press the **enter** key.
5. Press the **arrow** keys to raise or lower the specific conductance to match the calibration standard in mS/cm.
6. Press the **enter** key to finish calibration of specific conductance. If the unit rejects the calibration, the display will show "FAIL" before returning to the "Calib" screen.
7. Press the **Esc**  $\infty$  key to return to the real-time data screen. Record the final or post-calibration specific conductance readings into the logbook.
8. To check with a lower conductivity standard, repeat steps 1-3 with the lower standard.

#### B) Probe Maintenance

Clean the oval measurement cell on the specific conductance sensor with a small, non-abrasive brush or cotton swab. Soap or rubbing alcohol may be used to remove grease, oil, or biological material. Rinse with water.

**The Conductivity probe accuracy range is +/- 1% +/- 1 uS/cm . For example, a solution that is 1000 MicroSiemens, the range would be 1000 x 0.01 +/- 1 MicroSieman or 10 +/- 1 MicroSieman.**

#### 4) pH

##### A) Probe Calibration (a two-point style calibration)

1. Rinse the probe 3 times with deionized water.
2. Rinse the probe 2 times with a small amount of the 7.0 pH standard.
3. Fill cup with 7.0 pH standard to within a centimeter of the top of the cup. Wait for the readings to stabilize. Record initial or pre-calibration specific conductance readings in the logbook.

4. Press either of the **arrow** keys to cause the "Calib" icon to blink instead of "Screen". Press the **enter** key to select calibration. Use the **arrow** keys to cause "pH" to blink and then press the **enter** key.
5. Press the **arrow** keys to raise or lower the pH to match the calibration standard.
6. Press the **enter** key to finish calibration of pH. If the unit rejects the calibration, the display will show "FAIL" before returning to the "Calib" screen
7. Press the **Esc**  $\infty$  key to return to the real-time data screen. Record the final or post-calibration pH readings into the logbook.
8. Repeat steps 1-7 for the second pH standard. This pH standard will depend on the types of streams that will be encountered. Use 4.0 pH if mainly acid streams will be encountered and use 10.0 pH if mainly alkaline streams will be encountered.
9. When finished with the second pH standard, add a very small amount of tap water (just enough to keep the air inside the cup moist) to the storage cup. **THE STORAGE WATER SHOULD NOT COVER THE PH OR DO PROBE WHEN THE SONDE IS HORIZONTAL.**

#### B) Probe Maintenance

Two electrodes are used to measure pH: a glass pH probe and a reference electrode enclosed in a reference sleeve. If the response time for pH seems slow, refer the owner's manual for cleaning instructions.

**Glass pH probe:** Little maintenance is required. Check the tip of the probe to make sure the glass is not broken or dirty. If the pH sensor is obviously coated with oil, sediment, or biological growth, clean the glass with a very clean, soft, non-scratching cloth wet with rubbing alcohol (a cotton ball will do). Rinse with tap water.

**Reference electrode:** Gently pull the reference sleeve away from the probe. The reference sleeve is the black tube with a porous Teflon Reference Junction attached. Discard the old electrolyte from the reference sleeve. Refill the sleeve to the top with reference electrolyte. With the probe pointed toward the floor, push the full reference sleeve back onto its mount until the sleeve has just covered the first o-ring located on the mount (just behind the silver electrode). Turn the probe so that the sensors point toward the ceiling and push the sleeve the rest of the way onto its mount. Rinse with tap water. The porous Teflon

Reference Junction is the most important part of the pH performance. Make sure it is clean and passes electrolyte readily. If not, replace it. When seating the reference sleeve, trapped air and excess electrolyte is purged. This purging flushes and cleans the porous Teflon Reference Junction.

**The pH probe accuracy range is +/- 0.2 pH units (i.e., 6.8-7.2 is an acceptable reading for 7.0 buffer solution.**

#### **5) Temperature**

**The Temperature probe accuracy range is +/- 0.2<sup>o</sup> C.**

#### ***Quanta G Probe Storage***

When not in use, the H<sub>2</sub>O should be stored with the storage cup containing about ½ inch of tap water. In an emergency, the cup can be filled with ½ inch of clean creek water. The creek water should be replaced with tap water when you return to the lab. The pH reference electrode should also be stored in saturated KCl solution under the plastic cap.

### **QUALITY ASSURANCE/QUALITY CONTROL**

Calibration logbooks are maintained for each instrument. Any instrument failing to meet calibration requirements is repaired or shipped to the manufacturer. Meters are calibrated weekly, prior to sampling and are recalibrated in the field, if conditions warrant. For example, if a Hydrolab has been calibrated for pH using the 7 and 10 buffers, recalibration is performed if a stream pH of 3 is encountered.

Each meter has an identification number, which is recorded on the habitat assessment sheet each time the meter is used. Should any instrument fail to calibrate, readings taken prior to the failed calibration may be erroneous. Documentation of the instrument used at each site will help to keep data loss to a minimum.



## SOP B. INSTRUCTIONS FOR COMPLETING THE HABITAT EVALUATION FORM

### DESCRIPTION OF STREAM ASSESSMENT PARAMETERS

The quality and quantity of habitat is a major determinant of aquatic community potential. Consequently, a thorough habitat characterization is essential for proper interpretation of biological assessment results.

The following is a discussion of the stream assessment parameters outlined on the WVDEP WAP Stream Assessment Field Form. It is intended to provide information on interpreting each parameter as well as identifying the value(s) of resultant data.

#### I. INITIAL SITE SURVEY

A WAP field crew consists of two individuals charged with collecting habitat and biological/physicochemical data. Throughout the following discussions, the term "Geomorph" will be used to describe the crewmember in charge of collecting habitat information. "Biomorph" is the term used to describe the crewmember in charge of collecting biological and physicochemical data.

USGS topographic maps with a 1:24,000 scale will be used to navigate to sampling sites (GIS or Geographic Information System maps on Laptop, County Maps, or Gazetteer Maps are supplemental). The map coordinator should have marked all sites (pink for random sites, yellow for target sites) before sampling begins. After the location of the stream site has been confirmed, the Geomorph is responsible for establishing a 100-meter assessment area. The Geomorph will actively traverse the stream from one end to the other taking note of pertinent habitat information and measuring the 100 m reach. **NOTE: the Geomorph will avoid walking in the stream until physicochemical samples have been collected and avoid stepping in riffles that may be used in macroinvertebrate and periphyton sampling. "THERE SHOULD BE NO DEVIATION FROM THE ABOVE PROTOCOL. THE GEOMORPH MUST COVER THE ENTIRE 100 m STREAM REACH TO ACCURATELY COMPLETE THE HABITAT FORM. THIS CANNOT BE DONE STANDING AT ONE END OF THE REACH OR FROM THE VEHICLE."** The Geomorph will perform other duties concurrent with the establishment of the 100 m assessment reach (outlined later in detail). Procedures specific to each sample type are discussed below.

## II. ACCESSING THE SITE

Due to the remoteness of some sites (usually reference and random), traversing to the sample site may require long strenuous hikes over difficult terrain (NOT DANGEROUS TERRAIN!). If a long hike is necessary to get to a site, carefully consider the terrain and your personal ability and health to access the site. If you feel it is too difficult (e.g., too far to hike or too deep to wade) or dangerous (e.g., steep banks) to get to the site or assess it, do not attempt it. Discuss it with other WAP teams who may be willing to try to get the site later. **DO NOT NAVIGATE TO ANY ASSESSMENT SITE THAT PRESENTS A DANGEROUS SITUATION TO YOU OR ANOTHER TEAM MEMBER.**

### A. Random Sites (EPA Probabilistic Sites)

*An attempt should be made to access random sites no matter how far the hike unless it appears dangerous or too difficult to do so.*

Beginning in 2002 the Random Sampling Program switched from a watershed specific sampling effort to a statewide effort. The state has been divided into 25 watersheds by combining smaller watersheds with the larger ones as follows:

1. Potomac Direct Drains and Shenandoah (Jefferson County)
2. Cacapon and Shenandoah (Hardy County)
3. Cheat and Youghiogheny
4. Upper New and James
5. Lower Ohio and Big Sandy
6. Monongahela Direct Drains and Dunkard Creek

Six target to eight sites in each of the 25 watersheds must be fully sampled for water quality, benthos, periphyton, and habitat each year. Additionally, we will be conducting substrate characterization studies (or "pebble counts") at one-third of the sites. Target sites are defined as riffle/run habitat, wadeable, and can be sampled using kick protocols that result in comparable data.

The site list for each watershed will consist of at least twelve samples. See the following pages for an example of a site list. The first six to eight are the sites designated for the given year. The remaining six are backup sites to replace any sites in the first six that are not target sites or inaccessible due to landowner access denial or extreme physical barriers.

Since you know you will be visiting the first six sites, they may be sampled in any order. This will allow you to work more efficiently, as some sites may not be adjacent on the list, but are actually nearby. However, **you must do the pebble counts at the first two sites on the list.** Unlike duplicate sampling

there can be no site substitutions, as this will bias the final analysis of the results. If you are unable to sample one of the first two streams (*i.e.*, the sites are not target sites or access was unavailable) the pebble count must be done at the third stream on the list.

For example (Referring to Figure B-1): If you were working the stream list from the mouth up, you might sample Hog Jowls Run and Badgely Fork first, since they are near the mouth and close together. When you get to Sang Run, the landowner will not allow access, so you can't get a pebble count. You must then go back to Hog Jowls Run and do a pebble count there. If Hog Jowls wasn't acceptable as a target site, then it should be done at Job Run, etc. on down the list.

Backup sites **MUST** be sampled in the order presented on the list, since you will be replacing these on a one-on-one basis.

The little random maps (the ones with the hexagonal cells and site plot) will no longer be provided as of 2002. Instead, the coordinates are included in the stream list. Sites are still plotted out on topographical maps. In addition, an ArcView coverage of the sites will be provided for use on the field laptops.

R#	ANCODE	STREAM NAME	Latitude	Longitude	STR_ORD	TOPONAME
<b>DO THE FOLLOWING SITES IN ANY ORDER</b>						
R#2008	WVKC-39-{2.4}	Sang Run	38 41	0.42 81	9	25.99 1 Tariff
<b>DO PEBBLE COUNT HERE</b>						
R#2010	WVK-34-{3200}	SPRING CR	38 51	22.11 81	20	15.18 4 Spencer
<b>DO PEBBLE COUNT HERE</b>						
R#2085	WVK-46-B-{1.2}	Hog Jowls Run	39 5	2.40 81	8	11.44 1 MacFarlan
R#2088	WVKC-10-P-1-A-{2.1}	Job Run	38 56	34.54 80	57	45.72 1 Tanner
R#2104	WVKC-10-T-15-A-{1.8}	Badgley Fork	39 11	0.59 81	32	43.58 2 South Parkersburg
R#2137	WVKC-31-G-{1.9}	McGregor Run	39 18	44.41 81	1	52.57 1 Ellenboro
<b>THE SITES BELOW ARE BACKUPS AND MUST BE DONE IN THE ORDER ON THIS LIST.</b>						
R#2157	WVPC-12-{1.2}	Sandy Creek	38 45	4.17 80	18	12.35 1 Rock Cave
R#2170	WVPC-24-{1.7}	Rowles Run	38 53	7.67 81	11	31.20 2 Annamoriah
R#2237	WVP-19-D-{4.9}	Swollen Knuckles Ck	39 14	39.26 81	1	51.92 1 Harrisville
R#2260	WVS-11-{2.5}	Straight Fork	38 43	41.24 80	47	31.85 1 Gassaway
R#2355	WVP-19-E-2-{0.1}	Loveberry Run	38 59	21.91 80	35	19.97 1 Peterson
R#2356	WVPC-7-3-{0.1}	Mill Fork	38 41	45.58 80	58	55.70 1 Rosedale

Figure B-1. An example of a typical Random Site List

### *Locating the X-Site*

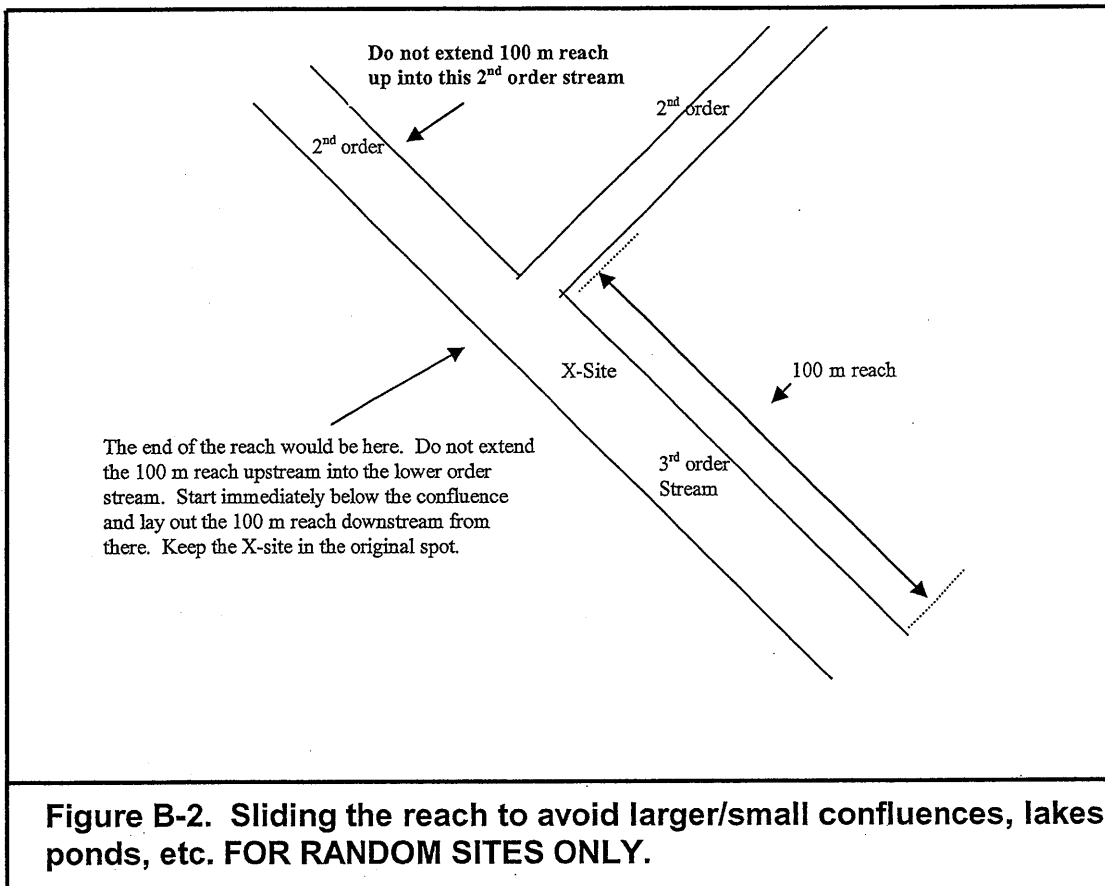
USGS topographic maps with a 1:24,000 scale are marked with an **X (highlighted in pink)** to designate the sampling station for random sites. This spot is referred to as the **X-site** and is the downstream end of a 100 m reach that is to be assessed (some situations require sliding the reach and thus the X is not at the downstream end; see below for details). **NOTE: Always collect physicochemical samples and GPS coordinates at the X-site for random stations. If possible, get coordinates from the stream channel and let the GPS run for several minutes (5-10) before recording the latitude and longitude.** WAP teams should use all available means to ensure that they are at the correct location; including Laptop GIS programs, topographic, county, and/or gazetteer maps. GPS units should also be used to confirm the X-site latitude and longitude that is provided on the EPA sheet for each random station. If the GPS coordinates and the given X-site coordinates differ by more than 10 seconds, re-check your position. **NOTE: You should make an attempt to get an exact match if possible.** There will be stations where the GPS unit will not track satellite vehicles (SV's) and thus confirmation of the X-site coordinates may be impossible. Team members should collaborate in these instances and utilize their best professional judgment (BPJ) to decide where the X-site is located (finely tuned map reading skills are important).

After the X-site has been confirmed (or located via best professional judgment), the Geomorph will establish a 100-meter assessment area. If there are no riffle/run habitats within 100 m reach, the site is considered non-target for random sites and should not be sampled. **Note: For random sites, our target stream has riffle/run habitat, is wadeable, and can be sampled using kick protocols that result in comparable data.** If you are denied access to a site either by landowners (*i.e.*, direct verbal communication or by best professional judgment that you should not ignore posted signs or fences) or by physical barriers (not gates or fences, but natural obstacles that involve dangerous conditions like steep gorges, forest fires, or floods), classify the site as target or not based on best professional judgment and clues that may be gathered about the stream. A good example is an agriculture stream where you are denied permission to the site but can see it well enough to properly classify it. If you cannot see the site, use GIS data, information from locals, what you know about other streams nearby, and what you can gather about the stream from other accessible points up or downstream.

### *Sliding the Reach*

There are some conditions that may require "sliding" the 100 m stream reach around features we do not wish to sample across. Do not proceed upstream into a lower order stream or downstream into a larger order stream when laying out the stream

reach. The map coordinator will note on the stream list any random 100 m reach that might require sliding due to the confluence of streams. If such confluence is encountered, note the distance and mark the confluence as the reach end. Make up for the loss of the reach length by sliding the other end of the reach an equivalent distance away from the X-site, as shown in **Figure B-1**. **NOTE: the confluence must be within the initial 100 m reach for this sliding to apply.** Do not slide the reach to avoid human disturbances like bridges, culverts, rip/rap, or channelized areas. If you have to slide the reach, make sure it is documented on the stream assessment form and include why it was moved and where. Include this information in the sketch of the assessment area.



Additionally, if the reach contains a lake, reservoir, or pond, mark the water body as the reach end and make up for the loss of the reach length by moving the other end of the reach an equivalent distance from the X-site (**See Figure B-2**). However, if the X-site is completely within a lake, pond, or valley fill, no sampling can occur and only the front page of the habitat form needs to be filled out describing the situation thoroughly. It is important to describe in detail on the assessment form any deviations from the standard layout.

In order to determine the stream reach, the Geomorph will actively traverse the stream (NOTE: the Geomorph will avoid walking in the stream until physicochemical samples have been collected and avoid stepping in riffles that may be used in macroinvertebrate and periphyton sampling) from one end to the other taking note of pertinent habitat information and measuring the 100 m area. The Geomorph will perform other duties concurrent with the establishment of the 100 m assessment area (outlined below in detail). Random sites have specific requirements for physicochemical sampling. The list of parameters that must be collected at all random sites can be found under SOP D and on the WAP "CHEAT SHEET".

### **B. Target Sites**

*Target sites should be assessed if they are less than one mile (or at crew members discretion if further away) from the vehicle unless it appears dangerous or too difficult to do so.*

USGS topographic maps with a 1:24,000 scale are marked with an **X (highlighted in yellow)** to designate the sampling station for target sites. If possible, the assessment reach should be established above bridges. Additionally, bridges should not be included in the assessment reach, if possible. Target sites include high quality, severely impaired, moderately impaired, non-impaired, unassessed, and 303d listed streams. These sites differ from random sites as indicated by the following:

- 1) There are no predetermined coordinates for the X-site unless otherwise noted on the stream list. The latitude and longitude will be determined after the sample site has been chosen.
- 2) There is more latitude in making decisions on where to conduct the stream assessment.
- 3) Latitude and longitude (coordinates) should be recorded at the downstream terminus of the 100 m assessment reach at all times (sliding the reach not applicable).
- 4) In general, all streams are sampled at the first readily accessible riffle/run upstream from the mouth.
- 5) Assessments are conducted as close to the mouth of the stream as possible, unless the stream is being sampled at multiple points.
- 6) Assessments are conducted upstream of road bridges/culverts if possible.
- 7) Physicochemical samples are collected at the downstream terminus of the 100 m reach.

**It is important to keep in mind that riffle/run sites are preferable to MACS sites when it comes time to report data as they are more abundant and only riffle/run**

data can be used to calculate a comparable WVSCI score. For example, if a riffle/run site can be found a ¼ mile further upstream without going above a significant tributary or changing land use (agriculture, etc), then go and sample the riffle/run site. **NOTE:** If a site is moved from the location marked on the map then a form should be filled out for both locations; the original site will have a front sheet detailing why it was unsuitable, and the new site will have a regular form pointing to the original site form and that it was moved. In general, do not collect a MACS sample unless the stream list indicates that the site is of special concern and should be sample regardless of the habitat type present. Describe in detail the type of MACS habitat present in case a future visit is scheduled.

Some conditions may require establishing the stream reach around features we do not wish to sample across. Do not establish a 100 m reach that includes a nasty discharge (e.g., AMD tributary, point source outfall, etc.). If a water quality impaired tributary is encountered within the chosen stream reach, move above the confluence a short distance and reestablish the 100 m reach and perform all WAP protocols. Additionally, fill out a form and collect appropriate physicochemical samples downstream of the confluence and from the mouth of the polluted tributary or outfall/source. If the nasty tributary is not on your stream list or the stream list for other WAP crews, conduct a full WAP assessment on the nasty tributary. **Provide detailed notes and document the specifics of the assessments and samples collected for all of the above.**

There is no generic list of physicochemical parameters for target sites other than field readings (Hydrolab/YSI) and fecal coliform bacteria. Sampling for specific parameters is determined on-site and is based on the surrounding land usage (*i.e.*, total phosphorus in agricultural areas when a problem is suspected, or metals in areas of mining). GIS software and coverages on laptops detailing the land use of each stream will be provided to the team with the topographic maps and stream list. These maps should be consulted to provide insight as to what parameters should be measured at the site. Another important way to get information about the land use is to observe what is in the watershed if you pass through it on the way to the site or the next site.

In some instances, a stream may appear to have an excellent water quality and habitat upstream of the targeted site. If this is the case, make all attempts to sample the segment that is a potential reference site or make notes about the stream segment and report it to other WAP teams and personnel to determine if it is a possible reference site candidate later (see the section on Reference Sites below).



### ***C. Duplicate Sites***

In order to fulfill quality control requirements (QC), a select number of duplicate sites will be assessed in each watershed. The letters **DUP 1** or **DUP 2** will follow the stream name on the stream list given to each WAP team. **NOTE: the stream listed is only a randomly picked site at which to complete a duplicate.** A duplicate can be performed at any site that meets certain needs. The assessment area should contain a large enough riffle/run area to obtain two complete benthic macroinvertebrate samples without any overlap (4 kicks versus 4 kicks). Make sure the instream substrate & velocity of the benthic sampling sites are as similar as possible. If the stream does not have an adequate amount of riffle/run habitat to collect two full samples, it will be necessary to substitute a replacement at the next stream that does have adequate habitat. If the first site you visit on a list provides enough good habitat to do a duplicate, then sample it as a duplicate. Do not wait until the end of a week or list to sample for a duplicate stream. Both team members will complete the habitat forms, collect benthic macroinvertebrates, and obtain appropriate physicochemical samples as if they are the only person there. **DO NOT PUT YOUR BENTHIC SAMPLING DATA ON THE OTHER PERSONS FORM.** Hydrolab/YSI and flow readings should be recorded on the DUP 1 assessment form only. GPS coordinates can be shared. **Make sure the name of the collector (not both team members) is written on the sample containers as well as a “-Dup 1” or “-Dup 2” at the end of the AN-code.** If the names of both team members are written on the containers there will be no way of determining the actual collector and thus no way of comparing the results for quality. If for some reason the designated duplicate is not sampleable, the team should replace the duplicate site with another stream in the same week.

### ***D. Alternate Sites***

Alternate sites serve as replacements for target sites that have been judged non-sampleable (streams that are dry, not wadeable, or are inaccessible). The map coordinator will provide a list of alternate streams (designated by **ALT** after stream name) to draw upon in the event an original target site is found non-sampleable. The alternate sites are classified using a system similar to target sites and include moderately impaired, non-impaired, high quality, and unassessed streams. Substitute all non-sampleable target sites with an alternate site of the same classification, (*i.e.*, replace a moderately impaired target site with a moderately impaired alternate site). If your list does not have an appropriately classified alternate, it may be necessary to collaborate with another team to select a replacement.

### ***E. Reference Sites and Potential Reference Sites***

*Potential reference sites and established reference sites should be assessed no matter how far the hike unless it appears dangerous or too difficult to do so.*

Reference conditions describe the characteristics of stream segments least impaired by human activities and are used to define attainable biological or habitat conditions.

**Therefore, it is extremely important for WAP teams to conduct assessments on several (as many as possible) undisturbed streams that meet reference conditions.** The map coordinator will provide each team with a list of potential reference sites and already established reference sites. Candidate reference sites were established by examining GIS land use coverages and marking the stream segments that appear to have the least amount of disturbance. Because the GIS coverages may not be current or complete, many of these candidate sites will not meet reference conditions and, thus, should not be assessed unless otherwise directed. On the stream list.

All potential reference sites and already established reference sites should be reconned by vehicle if not done so previously. The map coordinator will indicate on the stream list whether a candidate site has been previously reconnoitered. If a stream segment on your stream list has not been previously reconnoitered, refer to *Steps for determining candidate reference sites while in the field*, below.

**WAP teams should note that they are by no means limited to the list of potential reference sites provided by the map coordinator.** If a potential reference site is encountered while in the field, every effort should be made to conduct a full WAP assessment on that stream segment. If a potential reference site is also designated as a target site, then search for a place to sample that will satisfy potential reference conditions. In other words, if a small disturbance is encountered at or near the mouth of a stream that is not designated potential ref on the stream list, move the site above the disturbance to conduct the assessment.

**Always collect "RANDOM SITE" physicochemical parameters at all potential and established reference sites.**

Because of the nature of reference sites (undisturbed), traversing to the sample site may require long strenuous hikes over difficult terrain (NOT DANGEROUS TERRAIN). This should not be a reason for eliminating the site for assessment. If you personally feel it is too difficult (or too far to hike) to get to the site, do not attempt it. Discuss it with other WAP teams who may be willing to give it a try. **DO NOT NAVIGATE TO ANY ASSESSMENT SITE THAT PRESENTS A DANGEROUS SITUATION TO YOU OR ANOTHER TEAM MEMBER.**

Steps for determining candidate reference sites while in the field:

Determine human disturbances by reconnaissance and using GIS land use maps. Choose stream segments with no major (or as little as possible) human disturbance, (*i.e.*, eliminate sites with strip mines, refuse piles, towns, major roads, active open fields or agriculture), impoundments, power-lines, non-point sources, etc. **Consult the GIS land use maps for determination of upstream disturbances.** Some of these disturbances are indicated on topographic maps. If possible choose candidate sites located within a State Park or other static land use type. In most cases, it will be necessary to choose candidate sites with limited accessibility (obviously due to the nature of the condition we are searching for) that requires some long hikes. If passable jeep trails or hiking trails are indicated on the map, try and choose sites within their paths and make the hiking distance as short as possible.

Anthropogenic disturbances within the stream assessment area should be evaluated visually. Best professional judgment is employed to make reference site inclusions based on the number and type of disturbance. For example, a surface mine site would generally be considered a greater disturbance than an ATV trail and small road combined and would exclude the site from reference condition consideration. However, impacts from the ATV trail and/or road may be considered minor so that they do not exclude the site from reference consideration. In particular, don't immediately eliminate a site as potential reference if it has a small road following along much of its length unless there is obvious erosion or areas of high sediment deposition. Many of our established reference sites do have roads running parallel to them or crossing them at some point(s). Also, consider where you are in the state when deciding on potential reference sites. The northwestern portion of West Virginia (Western Allegheny Plateau – Ecoregion 70) should not be held to the same standard as the eastern mountainous section (Ridge and Valley – Ecoregion 67). In other words, the least disturbed conditions in Ecoregion 70 are not equal to those of Ecoregion 67. For example, some streams in the Upper Ohio South watershed in Ecoregion 70 have hilltop farms that may offer little if any impact to the stream located a down in the valley below. Some of these are established reference sites and represent the best possible conditions for the Ecoregion. In Ecoregion 67, there are many streams without any recent land disturbance (entirely forested). Many of these are established reference sites.

The following selection criteria are used to select reference sites after assessments have been conducted and the data entered in a database. It will be impossible to utilize all of them while in the field. However, it will be useful to consider these criteria while making decisions on whether to conduct an assessment on a candidate reference site. A stream assessment form will be necessary when considering the criteria.

**\* Indicates criterion that can be determined in the field.**

1. D.O. > 5.0mg/l \*
2. pH between 6.0 and 9.0 \*
3. Conductivity < 500 µmhos/cm
4. Fecal coliform < 800 colony/100ml
5. No violations of State WQ Standards
6. No obvious sources of non-point pollution \*
7. Epifaunal substrate / available cover score >10\*
8. Channel alteration score >10 \*
9. Sediment deposition score >10 \*
10. Bank vegetative protection score of lowest side >5 (right & left banks ) \*
11. Undisturbed vegetation zone width score of lowest side >5 (right and left bank scored separately) \*
12. Total habitat score > or = 130 points \*
13. Evaluation of anthropogenic activities and disturbances (see page 2 of Stream Assessment Form) \*
14. No known point source discharges upstream and within view of assessment site (completed after 1-13 are met)

It must be understood that absolute pristine habitat conditions do not exist in most areas. Therefore, decisions must be made on what is an acceptable level of disturbance to represent reference condition. Additionally, acceptable conditions may differ among watershed regions because of factors such as local geology, vegetation, and predominant land use. In heavily disturbed watershed regions, undisturbed conditions may not exist. Consequently, reference conditions may need to be determined based on the best available conditions. In some cases, a reference site may simply not exist in that watershed and reference sites from other watersheds will have to be sampled instead.

These selection criteria were developed to obtain reference conditions for streams that were assessed in the 1996 and 1997 field seasons. The assessments covered 12 watersheds and over 1,000 streams were assessed. Most assessments were performed at the mouth or as close to the mouth as possible. Generally, no effort was made to select candidate reference sites before assessments began. Priority was given to impaired streams and/or streams currently on the 303d list. Small subsets of unassessed and unimpaired streams were also visited. During the 1997 field season, approximately 150 probabilistic sites were assessed. This placed heavy demands on the field personnel and generally prohibited efforts to locate and assess candidate reference streams. Field crews were instructed to note all outstanding streams that were encountered during the targeted or random assessments and mark them as possible reference sites on a stream assessment form. Plans were implemented to establish a list of candidate reference sites prior to the 1998 field season and time was allocated to ensure that they are visited.

The criterion for dissolved oxygen was taken from WV Water Quality Standards as developed by the State Water Resources Board (SWRB). The criterion for conductivity was established from observations (no statistical work) of WAP and DWWM (Division of Water and Waste Management) data and from the best professional judgment of several experienced field employees. The fecal coliform value of 800 colonies/100ml is double the maximum set by the SWRB, which states that fecal coliform shall not exceed 400 colonies/100ml in more than 10 percent of all samples taken during the month. This value was raised to 800 colonies/100ml for reference criteria due to the lengthy holding times of fecal samples (24 hours in many cases). Additionally, experienced field personnel have encountered fecal counts exceeding the standard in streams where no human impacts were apparent. Thus, a value of 800 colonies/100ml would decrease the possibility of excluding some undisturbed (anthropogenically) streams from reference consideration.

Criteria 7 through 12 are adapted from the RBP habitat assessment modified for use in the USEPA/EMAP program. These criteria were selected because they are presumably most indicative of anthropogenic perturbation. A value  $> 10$  indicates that stream habitat is at least sub-optimal for that particular parameter. The WAP sampling strategy dictates that assessments be conducted at or near the mouths of streams. This strategy tends to bias the habitat scores (many sites are roadside accessible or below bridges) and in many cases results in relatively low scores for those parameters that are most indicative of human disturbance. It is for this reason that the minimum values are set to 11 (7 through 10) and 6 (parameter 11). Otherwise, few streams (if any) would meet the selection criteria.

### III. GUIDELINES FOR COMPLETING THE STREAM ASSESSMENT FORM

#### Important Note

If a stream is dry or not sampled, Page 1 of this form must be completed. If water quality only is collected, you must complete pages 1-4, as best as you can.

#### FRONT SIDE OF ALL PAGES

REVIEWERS INITIALS: All habitat forms must be reviewed and initialed for completeness by a second crew member. This review must be performed on-site. The Biomorph should point out any omissions to his/her partner and initial the page when all the data are complete. In the case of duplicate sites, this is an opportunity for each field worker to discuss discrepancies between the forms. However, all results should be considered final and should not be changed to match the other person's results.

AN-CODE: It is extremely important that the **correct** AN-code be recorded on each sheet as this is one way to link all the sheets for a sample together if accidentally separated. See below for more details about an-codes.

Date: Use mm/dd/yyyy format: *e.g.*, 04/29/1999. It is extremely important that the date be recorded on each sheet as this is another way to link all the sheets for a sample together if accidentally separated.

#### PAGE 1

##### Stream Verification

Stream Name and Location Description: Make sure the stream name on the map corresponds with the assigned AN-code from your printed stream list. If they do not match, make a note of it on the habitat sheet and printed list. Include a detailed description of the location such as: Greenbrier River US (abbreviation for Upstream) of Big Run at Hilldale Bridge, New River DS (abbreviation for Downstream) Lick Run at Glen Lynn, Red Creek Between Oak Run and Pine Run at Laneville, Piney Creek Upstream Beckley PSD 50m, Pinnacle Creek DS right UNT 0.5 miles south of Pineville, Bear Run near mouth south of Sanoma Upstream first bridge, Camp Creek at mouth in Camp Creek St. Forest at Campsite #2, etc. Be sure to include the receiving stream in the name of any source discharges (*e.g.*, Beckley PSD outfall discharging into Piney Creek US of Smock Run).

AN-code: (Alpha-Numeric Code) It is extremely important that the **correct** AN-code be recorded for each stream site. Mistakes in translation from the printed stream list to the habitat sheet must be avoided. Mistakes in this step create mass confusion and plenty of extra work during data entry. All streams will have an AN-code with the mileage designated between brackets (*e.g.*, -{3.6}). If you are going to sample at a location other than those listed, create a unique AN-code such as KG-3-#{1}. The mileage can be assigned to this AN-code later using the WCMS (Watershed Characterization and Modeling System) GIS model by the field personnel or the map coordinator.

Date: Use mm/dd/yyyy format: *e.g.*, 04/29/66

Time: Use military time. *e.g.*, 1315

Geomorph: Initials of the team member completing the habitat form.

Biomorph: Initials of the team member collecting macroinvertebrate and water samples.

Basin: *e.g.*, Upper Kanawha, West Fork, Lower New

County: *e.g.*, Hardy, WV

Quad: Enter the topographic quadrangle name, *e.g.*, Cass, Mt. Nebo, and Panther

GPS #: A number identifies Each GPS unit. Record that number here. This procedure will allow a malfunctioning unit to be identified.

GPS Station #: If a Gamin unit is being used, record the word "Garmin". If using Trimble units, record the name (code) assigned to the stream site coordinates that were stored in the GPS unit. Use the proper AN-code without the WV and without the dashes. Also, if a point (.) is used in an AN-code please designate it as "PT" since the GPS will not allow the use of ".". If all 8 spaces allowed by the GPS are not used, begin entering the stream name. Since stream codes for the EPA probabilistic sites can be very long and complicated, the GPS station number should use the "Random Number" instead (ex. RANDO123 for random site 123). If the GPS unit cannot lock into enough satellites to obtain a reading, enter "no fix" for GPS station #. If a Garmin unit is used to obtain coordinates, fill in this blank with "GARMIN". If the WCMS GIS Software is used to determine the coordinates, indicate as such on the form.

Examples follow:

(1) Banner Hollow (WVK-65-D) would be stored in the GPS as **K65DBANN**

(2) Wolfpen Hollow (WVK-58-B.1) would be stored as **K58BPT1W**

(3) Paint Creek was sampled at Collins Branch (WVK-65-{4.8}) and Burnwell (WVK-65-{3.1}). **K654PT8P** would be used to designate the Collins Branch site and **K653PT1P** would be used to designate the Burnwell site.

(4) Random Sites – Follow this protocol for naming: “RANDO123”, “RANDOM34”, “RANDOM78”, etc.

EPE: Record from Garmin GPS after XY’s have been recorded.

Random #: EPA Probabilistic (Random) sites are designated by a special number. This number (which will be on the stream list or topo map) is entered here.

XY’s Proofed and By: The person that double-checks the coordinates for accuracy in the office and the type of basemaps used as a reference.

EPA or Corrected Latitude and Longitude at X-site: Either the coordinates provided on the stream list for EPA Probabilistic sites (randoms) are recorded here or corrected versions of the coordinates are recorded here in the office after they were proofed in the office (see the item above).

Field Latitude and Longitude: Enter for all sites after obtaining readings in the field using Garmin or Trimble GPS units.

X-site Field Verified? : Answer appropriately. **YES** or **NO**. This must be answered.

If no, why?: Sometimes it is possible a stream site will not be physically visited. This may be due to one of two things: Landowner access denial or a physical barrier. Landowner denial could come in the form of a verbal denial, which is absolute, or in the form of implied denial. Implied denial simply means that the crew has seen evidence that the property owner would not be agreeable to our presence in the stream and used best professional judgment to not sample the site. This evidence can come in the form of an abundance of posted signs (e.g., at every fence post), by conversation context talking to a neighbor (e.g., “He likes to shoot at trespassers.”), heavily fenced and secured areas, or simply a private property (e.g., the site is located in the back yard of a secluded cabin).

Is site target and kick sampleable?: Answer appropriately. **YES** or **NO**. This must be answered even if the site was not seen.

If no, why?: Sometimes a stream site will not be sampled for one reason or another. The following are possible reasons: (1) ephemeral, (2) low flow-permanent (non-drought, i.e., subsidence) or low flow-temporary (drought), (3) too deep-permanent (e.g., a larger stream or river that is flowing but always will be over the net) or too deep-temporary (e.g., a smaller stream that is over the net at that time but would not be at base flow), (4) no riffle/run habitat present, (5) filled, (6) impounded, (7) no stream present-map error, (8)



wetland—no defined channel, (9) other. If other reasons arise, please comment in sketch area on page 1 when appropriate. **ALWAYS FILL OUT THE FIRST PAGE OF THE HABITAT ASSESSMENT FORM AND GET COORDINATES OF THE SITE, REGARDLESS OF WHETHER ANY TYPE OF SAMPLING WAS CONDUCTED (EVEN IF STREAM IS DRY, IMPOUNDED, OR INACCESSIBLE)! THIS IS IMPORTANT INFORMATION AND ASSISTS IN DATABASE MANAGEMENT.**

Detailed notes on verification, access, and sampleability of site: Notes concerning the above four items and the process that led to the answers above.

Sample Type: Indicate which of the data types were collected (1) Hydrolab/YSI, (2) Lab water, (3) Fecal, (4) Habitat, (5) Bugs, (6) Periphyton, (7) Flow, (8) Pebble Count. **Do not include Hydrolab/YSI readings as part of the lab water data. This refers to laboratory-analyzed samples only.**

Sampled?: Answer appropriately. **YES** or **NO**. This must be answered. In some instances you may be sampling some aspect (e.g., WQ only) even if the site is declared to be non-target.

Sample Type: Indicate each type of sample taken: 1) YSI, 2) Lab Water, 3) Fecal, 4) Habitat (RBP Habitat), 5) Bugs, 6) Periphyton, 7) Flow, 8) Pebble Count.

WQ Sample Location: Indicate the cross-sectional location of the water quality sampling: 1) Mid-Stream, 2) Thalweg, 3) Left Bank, 4) Right Bank, 5) Other (please describe).

Hydrolab Method: Indicate the type of collection method used with the YSI/Hydrolab: 1) Grab, 2) Bucket, 3) Other (please describe).

Lab Water Method: Indicate the type of collection method used to obtain the lab water: 1) Grab, 2) Bucket, 3) Other (e.g., Cross-Section Composite; please describe).

Duplicate Type: If the site is assessed by each team member independently, the site is a duplicate site. **These sites should be treated as if each person was the only person assessing the site.** Indicate the type of duplicate it is 1) Lab Water, 2) Fecal, 3) Habitat, 4) Bugs, 5) Periphyton. Hydrolab/YSI readings should be recorded on the DUP 1 assessment form only. GPS coordinates can be shared. Make sure all sample containers are labeled with the person's name that made the collection, not both team members. This allows for tracking potential sampling errors resulting from poor technique or improper training.

Duplicate Number: The number designation of the duplicate sample, that is, Dup #1 or Dup #2.

Was site moved? [For Non-Random sites only]: Answer **YES** or **NO**.

Explanation? [For Non-Random sites only]: Explain why the site was moved and where the site was moved to. This may apply to random sites where sliding the reach is necessary. It can also apply to other sites that might be moved upstream or downstream from the point marked in order to obtain riffle/run habitat, etc. **If the site is moved, it is important to identify and mark the location of the new assessment site on a topo map with date and initials of team and fill out a form for both sites.**

Directions to Stream Site: Give a detailed description on how the stream site was accessed. Include highway names & numbers, distances from prominent landmarks (manmade and/or natural), proximity to towns, etc. Indicate if contact with landowner/stakeholder/groundskeepers, etc., are necessary and note where, when, and why they should be contacted. Addresses of and other specifics about the landowner/stakeholder/groundskeepers can be written down on page 8 under the section called Landowner/Stakeholder Information.

Bird's-eye-view Sketch of 100 meter Stream Assessment Area and General Comments:

Provide a detailed sketch of the area and include stream flow direction, stream morphology (i.e., riffles, runs, pools, bends, falls, large boulders), land use on left and right bank, upstream activities (if possible), proximity to permanent land marks, indicate direction by drawing a North arrow (↑), and any observations which may provide pertinent information to the assessment and location of the stream area. Indicate where GPS coordinates are collected by marking the spot in the stream with an **X**. **Coordinates should be obtained at the "EPA provided" latitude and longitude for random sites (usually downstream terminus). Coordinates should be obtained at the downstream terminus at all other sites if possible.** Indicate direction of flow with an arrow (↑). Mark the areas where benthic macroinvertebrates and periphyton are collected with a (b/p), and mark water sample collection areas with a (wq). Indicate the location of the preceding descriptive drawings within the 100 m assessment area and provide visual estimates of distance (try drawing it to scale). Indicate the upper end of the reach with an "us" and the downstream end with "ds" and attempt to correlate these with permanent landmarks. **Keep in mind that a different field crew may be revisiting the site in 5 years and will rely heavily on your description/drawing to get back to the same location. In other instances, it may be necessary to determine the location using GIS programs.** General comments can be very important when interpreting sample data. Therefore, any anomalies or outstanding attributes should be noted. If it is a random site and sliding the reach was necessary, indicate on the map the changes that were made and place an X in the drawing of the reach to indicate the X-site location.

Notes: General notes about the sample or sample location (e.g., This site is on a 303d listed stream, This site is taken at a previously sampled Gray WVSCI site, etc.).

**PAGE 2****Site Activities and Disturbances**

The information obtained from these measurements will aid in providing insight as to what organisms may be present or are expected to be present, and the presence of stream impacts. This information is also invaluable when conducting 305(b) assessments of streams.

Local Watershed Erosion: In the 100 m reach, note the **existing or potential** detachment of soil within the local watershed (that portion of the watershed that drains directly into the stream upstream of the sample point) and its movement into the stream. Indicate whether there is **none**, if erosion is **slight**, **moderate**, or **heavy**. Look for roads, drains, tilled ground, hillside slips, staging areas, etc. **Do not confine your observations to the local stream banks in the reach.** If observations are made outside of the upstream or downstream terminus of the 100 m reach, record them in the large "notes box" below.

Recent Stream Scouring: In the 100 m reach, note the **existing or potential** scouring of the substrate from recent high flow events. Look for scared or abraded substrate particles or the absence of periphyton in seemingly ok streams. Confer with the Biomorph after the first kick to determine if the benthos seems normal. Also consider other streams visited in the area. Information from locals can also be invaluable. If the stream does appear to be moderately or heavily scoured, confer with other crews or the office to determine if benthic sampling should continue or be postponed at the site.

Atmospheric Odors: Rate the any atmospheric odors based on the following scale: **none (0)**, **low (1)**, **moderate (2)**, **high (3)**, **extreme (4)**, or **not rated (NR)**.

Odor Description: Describe the nature of the odor

Local Watershed Nonpoint-Source Pollution: Refers to problems and potential problems **other than siltation** in the 100 m reach. Non-point source pollution is defined as runoff from agricultural lands and urban areas (e.g., shopping center parking lots). Other factors in a watershed that may affect water quality are feedlots, wetlands, septic systems, dams and impoundments, oily strips in center of roads, mine seepage, gob-pile runoff, quarry runoff, landfill leachate, wood-yard runoff and leachate, acid deposition, etc. Indicate **none**, **potential**, or **obvious sources**.

If obvious, magnitude?: If the Nonpoint-Source Pollution is obvious, indicate how intense it is by checking Slight, Moderate or Heavy.

Specify Obvious or Potential Sources of NPS: Indicate the obvious or potential source of NPS that you observed in the 100 m reach. Elaborate in the comments section if needed.

Point Source Discharges: Indicate the presence any pipes/culverts entering the streams within the 100 m reach? Indicate **Yes or No**.

Pt. Source(s): If there is a point source or sources located above the assessment reach describe it in the comments section.

Stream Assessment Area Activities & Disturbances: Record any of the following disturbances that were observed in the 100 m stream assessment area. Place a check in the box corresponding to each disturbance that is observed. Leave the box blank for any disturbance not observed. If one of the disturbances is observed above or below the 100 m reach, record it in the notes box below. The form is divided into the following major categories:

**RESIDENTIAL:** Note the presence of any of the listed residential disturbances adjacent to or near the stream.

**RECREATIONAL:** Record the presence of organized public or private parks, campgrounds, beaches, or other recreation areas around the stream assessment area. Look for evidence of informal areas of camping, swimming, or boating around the stream (e.g., swimming hole).

**AGRICULTURAL:** Note the presence of cropland, pasture, orchards, poultry, and/or livestock.

**INDUSTRIAL:** Record any industrial activity (e.g., chemical, pulp), commercial activity (stores, businesses) or logging/mining activities around the stream assessment area.

**MANAGEMENT:** Note any evidence of liming activity, water treatment, dredging or channelization, flow control structures, etc.

**TRANSPORTATION:** Record the road size as single, double, or multi-lane. Use best professional judgment to judge the size of roads. If you think two cars can pass one another without steering onto the shoulder, then designate the road as double lane. A single lane would require steering onto the shoulder to pass one another. Multi-lanes are large roads such as Interstate highways and some U.S. routes. Large industrial roads such as the ones built on strip mine operations may also be considered multi-lane. State and county maintained highways are usually roads that serve numerous purposes. If you cannot determine what the specific use of a road is this category will usually apply. It may also be helpful later on to write down a description of the road (e.g., haul-road, I-77, C.R. 52/3).

**HIGHWAY TYPE:** Record the road type as dirt, rutted dirt, applied stone (non-limestone), applied stone (limestone), asphalt, or concrete.

**Note** that the **RESIDENTIAL, RECREATIONAL, AGRICULTURAL,** and **INDUSTRIAL** categories each have a block for roads as well. Roads under these categories have specialized uses. For example, residential driveways and access roads to fishing sites (recreational), fields (agricultural), or mines (industrial). Using the key on the left side of the page, indicate the width of the road and the type of surface present.

**Comments Box:** “If known, what is the predominant land use(s) in this stream’s drainage? Is it mostly forested, agriculture, mining, logging, houses, urban? If mining present, is it active or abandoned, deep or strip, valley fills, etc. What is the predominant NPS of pollution? Are there point sources above the reach? Indicate if you used maps (GIS) or field verified comments. **DO NOT LEAVE THIS BOX BLANK!**”

This area is a good place to put comments about the land use observed from recon trips or gleaned from the GIS land use or topo maps. If comments are based on the map, note them as such. Landowner comments about the stream activities should also go here. The source of each bit of information should also be noted (e.g., WCMS, Topo, Recon, or Local or Landowner).

## PAGE 3

### Physical Characterization

**Average Stream Width (m):** Measure the wetted width of the stream at three transects representative of the 100 m assessment area. In general, the three measurements should be made at the downstream terminus, middle, and upstream terminus of the 100 m assessment area. These measurements will be used to calculate (40 x average width) the reach length for sites with substrate characterization scheduled. Streams greater than 30 m in width will require a visual estimate at three points following the above protocols (if stream conditions permit, try to get one actual reading). Record the measurements and calculate the average stream width. The **Geomorph** will take the measurements while establishing the 100 m assessment area (Note: do not walk in stream or take stream measurements until physicochemical data has been collected). A tape measure or measuring stick (thalweg pole) is provided for taking the measurements. The **Geomorph** must conduct this part of the assessment. The gathering of this information is important for several reasons. First, it provides data that is necessary to classify streams by size. Additionally, it requires the Geomorph to cover the entire 100 m reach that will allow for increased accuracy and consistency in the assessment of habitat.

Stream Rise Category and Estimate: Classify the stream gradient into one of three categories (low, moderate, or high) and estimate the stream rise in meters over the 100m stream reach.

Total Habitat Type % Coverage for Reach: Estimate the percent coverage of each habitat type (Riffle, Run, & Pool) for the 100m reach. **When considering the Pool coverage, remember to count biologically functional pools in smaller streams (i.e., do not use the <0.5 m cutoff used in the RBP).**

### **Sediment Characterization**

Sediment Odors: Disturb the sediment and note any odors described (include others not listed) which are associated with sediment in the area of the sampling station. Examine depositional areas for this parameter and collaborate with the Biomorph in making the decision.

Sediment Oils: Disturb the sediment and choose the term that best describes the relative amount of sediment oils observed in the stream sampling area. Examine depositional areas and collaborate with the Biomorph before making the decision.

Sediment Deposits: Note the deposits described (or include any other deposits not listed) which are present in the sampling area. Collaborate with the Biomorph before making the decision. Rate each sediment deposit as **0-None, 1- Low, 2- Moderate, 3-High, 4-Extreme, and NR-Not Rated (used if for some reason the substrate cannot be seen like when visiting a TMDL site during high turbidity events). Include the color of any metal hydroxides observed.**

Sediment Comments: Provided as a space to describe unusual substrates or qualities of the substrate. Use this area to elaborate on metal hydroxides (especially if more than one type is occurring simultaneously).

### **Substrate Particle Layer Profile**

At the X-site, randomly choose a location along the cross-section (Right, Middle, or Left depending on the seconds of the current time (0-20, 21-40, and 41-60 respectively). At this point, document the habitat type (Riffle, Run, Pool) and begin to remove and document the substrate layers (using the Substrate Size Classification outlined in Figure B-3) one at a time. If any sand or silt is documented, record the depth of that layer in cm. Repeat this until the top five layers are documented or until you reach the bottom of the biologically

inhabitable zone (no more than 15 cm). Record any notes that may be necessary. **Note:** The purpose of this evaluation is to document the colonization potential of the substrate relative to sedimentation. Therefore it is important to include Metal Hydroxides in the layer profile as they may have a smothering/cementing effect on the stream substrate in some situations.

**Figure B-3 Substrate Size Classification for Substrate Layer Profile and Dominant Substrate Type and Reach Characterization**

Class	Code	Size	Description
Bedrock	BR	>4000 mm	Bigger than car
Boulder	BL	>250-4000 mm	Basketball to car
Cobble	CB	>64-250 mm	Tennisball to basketball
Coarse Gravel	CG	>16-64 mm	Marble to Tennisball
Fine Gravel	FG	>2-16 mm	Ladybug to marble
Sand	SA	>0.06-2 mm	Gritty between fingers
Silt & Fines	ST	<0.06 mm	Smooth, not gritty (silt & muck)
Clay	CL	>4000 mm	Slick/ hard clay or hard-pan clay bottom
Metal Hydroxides	MH		Any Metal Hydroxide Deposit (Use this class only in the Substrate Layer Profile)

### Dominant Substrate Type and Reach Characterization

At various points along the stream reach where there is a sudden change in habitat type, substrate type, or both, measure the stream depth (m) using the thalweg pole and document the position (relative to the downstream end of the reach) along stream reach in meters, the habitat type (riffle, run, or pool), the 1<sup>st</sup> dominant substrate type (using the Substrate Size Classification outlined in Figure B-3) and its percent coverage and then the 2<sup>nd</sup> dominant substrate type and percent coverage. Take measurements throughout the reach (Geomorph responsibility). Some streams sites may not have all of the three habitat types (usually pools are missing). **When considering the Pool areas, remember to count biologically functional pools in smaller streams (i.e., do not use the <0.5 m cutoff used in the RBP).** *Note: Do not use Metal Hydroxides as a class when evaluating the dominant substrate type; look only at the functional size classes (i.e., those that have size ranges).*

## PAGE 4

### Field Water Quality Measures

**Flag:** Indicate if one of the recorded values was not accurate or suspected of being in error. This field may also be marked in by the data entry person (in pen) if they suspect inaccuracy of the instrument readings.

Physicochemical Parameters - temperature, pH, D.O., conductivity: Record the values for each of the physicochemical parameters indicated from the water probe. 1) Temp-°C, 2) pH-Standard Units, 3) D.O.-mg/l, 4) Conductivity-µmhos/cm.

Hydrolab/YSI I.D.: Record the instrument identification number.

Seasonal Water Level: Indicate the water level relative to the season as **(1) above normal, (2) normal, (3) below normal, or (4) Flooding**. **Example**: in general, high water in autumn would be above normal.

Water Odors: Record the odors described (include any odors not listed) that are associated with water in the sampling area: **(1) normal, (2) sewage, (3) petroleum, (4) chemical, (5) anaerobic (septic), (6) none, (7) other**. Collaborate with the Biomorph in making the decision.

Foam/Suds: Rate the any Foam or Suds on the surface of the water based on the following scale: **none (0), low (1), moderate (2), high (3), extreme (4), or not rated (NR)**.

Surface Oils: Note the term(s) that best describes the relative amount of water surface oils present: **(1) none, (2) flecks, (3) sheen, (4) globs, (5) slick**. Collaborate with the Biomorph in making the decision.

Turbidity: Indicate the term that best describes the amount of material suspended in the water column: **(1) clear, (2) slightly turbid, (3) moderately turbid, (4) highly turbid (or turbid)**.

Water Color: Indicate whether water color is normal (clear) or colored (e.g., orange for iron impacted streams).

Precipitation Status: Describe **precipitation events only** for the area during the time of visit and within the last 24 hours if possible. Comment on any heavy rainfall events, snowmelt, or storms that might have an impact on the water quality during sampling.

Peak Runoff: If it is raining or has rained recently, which of the following best describes the peak runoff (flush) condition of the stream at the site when water samples were collected: **1) <1 hour, 2) 1-4 hours, 3) 4 -12 hours, 4) 12-24 hours, 5) >24 hours, 6) Unknown**.

Stream Level: Indicate if the stream level is **1) at Baseflow, 2) Rising, or 3) Falling**.

## **Stream Bank & Riparian Vegetation Buffer Zone Measures**



Average Width of Intact Riparian Vegetative Zone (m): Check the box if greater than 50m for each side. If not, estimate the width of the intact (big tree, small trees, herbaceous layer) of each side.

#### Riparian Vegetation Classification:

This segment of the stream assessment form was developed to address certain objectives proposed in WAP's application for funding under the Wetland's Development Grant Program, 104(b)(3). The principal objective of the project is to assess the integrity of riparian vegetation zones in selected priority watersheds. The following parameters were indicated as possible measures for meeting the proposed objective:

- 1) Erodibility of riverbank soils
- 2) Density of bank vegetative cover
- 3) Riparian disruptive pressure
- 4) Riparian zone width
- 5) Percent trees, shrubs, herbs, (bank and riparian zone)

STREAM BANK VEGETATION performs a vital role in the control of erosion to streams. Trees and woody shrubs exhibit deeper and more permanent root systems than grasses and herbaceous plants and are, thus, more effective in reducing erosion throughout the year.

THE RIPARIAN VEGETATIVE ZONE serves as a buffer zone to pollutants that may enter a stream through runoff, controls erosion, and provides stream habitat and nutrient input into the stream. Relatively undisturbed riparian zones with large dominant tree species reflect healthy stream systems and are generally considered indicative of the best possible conditions.

The following visual estimation procedures are a semi-quantitative evaluation of the type and amount of different types of stream bank and riparian vegetation. The assessment will be used to evaluate the health and level of disturbance of the stream corridor.

The following discussion applies only to the Stream Bank / Riparian Buffer Zone Vegetation / Cover Type section on **Page 4** of the Stream Assessment Form.

While standing in a position perpendicular to the stream, visually establish a distance of **18 meters** from the right and left stream edge. This 18 m zone (one on each side of stream) will run parallel with the stream throughout the entire 100 m assessment area. Aerial coverage (described below) of the vegetation types will be conducted within this 18 m zone.

Determine the dominant vegetation type within the 100 m reach as 1) **Deciduous**, 2) **Coniferous (pines, spruces, hemlocks, cedars)**, or 3) **Mixed (>10%)**. Determination is made by considering both banks together.

Conceptually divide the stream bank and riparian vegetation into three layers: the **CANOPY** layer (> 15 ft high or 5 m), the **UNDERSTORY** layer (1.5 to 15 ft high or 0.5 to 5 m), and the **GROUND COVER** layer (< 1.5 ft high or < 0.5 m). Note that more than one vegetation type (e.g. grasses or woody shrubs) can potentially occur in more than one layer. Right and left banks are scored separately while looking downstream. Also, indicate the percent of **BARREN OR BARE SOIL** within the same 100 m reach and 18 m zone. This refers to highly erodable surfaces and does not include rock cliff faces or asphalt/concrete roads.

*The **CANOPY** category includes big trees such as sycamore, silver maple, box elder, river birch, cottonwood, and hemlock. The **UNDERSTORY** layer includes small trees and shrubby vegetation such as willow, alder, rhododendron, knotweed, wingstem, and multiflora rose. **GROUND COVER** vegetation includes ferns, mosses, and grasses.*

Estimate the aerial cover separately in each of the three layers. **The aerial cover can be thought of as the amount of shadow provided by a particular layer.** The maximum cover in each layer is 100%, so the sum of the aerial covers for the combined three layers could add up to 300%. The four entry choices for aerial cover within each of the three vegetation layers are "0" (absent= zero cover), "1" (sparse= <10%), "2" (moderate= 10-40%), "3" (heavy= 40-75%), "4" (very heavy= >75%). These ranges are provided as a key on the Stream Assessment Form.

When rating vegetation cover types, mixtures of two or more subdominant classes might all be given sparse ("1") moderate ("2") or heavy ("3") rankings. A very heavy cover class with no clear subdominant class might be ranked "4" with all the remaining classes either moderate ("2"), sparse ("1") or absent ("0"). Two heavy classes with 40-75% cover can be both ranked "3".

Stream Surface Shading (%): Stream surface shading plays a significant role in maintaining water quality in streams. Exposed streams will often experience increased water temperatures that may be directly or indirectly limiting to some organisms and may be favorable to nuisance algae and result in decreased dissolved oxygen. Light intensity may be favorable to some organisms and limiting to others. In general, a partially shaded (50-75%) stream achieves the greatest diversity. A fully shaded stream may inhibit the growth and reproduction of herbaceous aquatic and riparian plants. This situation can potentially inhibit primary production, cover, and habitat. However, this situation does provide better temperature control and increased allochthonous (organic material from outside sources) food resources.

Estimate the percent of stream surface shading using the following categories: fully exposed (0-25%), partially shaded (25-50%), partially exposed (50-75%), and fully shaded (75-100). Evaluate the shading based on a cloudless day in the summer at noon.

Riparian Vegetation Comments Box: *Describe your impression of the condition of the riparian zone in the 100 m stream reach. What is its' buffering ability? How intact is the riparian vegetation? Describe the vegetation species assemblage for both sides. Indicate the presence of human activities. Note the land cover type(s) immediately adjacent to the 18 m riparian vegetative zone on both left and right banks. Again, comments in this section are useful during 305(b) stream assessments.*

Amphibian Pool Present in riparian area?: Indicate if any of the following amphibian habitat types were present in the riparian area of the stream assessment reach:

- 1) **vernal pools** - vernal pools are an extremely scarce wetland habitat type occurring only where certain soil conditions are present. In late summer, fall and early winter, vernal pools appear as dry, dusty indentations mostly devoid of vegetation. Look for depressions filled with water along the stream bank and riparian zone.
- 2) **mud puddle** – small depressions in dirt roads.
- 3) **sediment ponds** - sediment ponds are built to trap runoff water. Sediment settles to the bottom of these ponds rather than accumulating in local creeks and streams. Typically found below valley fills and other mined areas.
- 4) **farm pond** – livestock watering hole or used for irrigation to crops.
- 5) **ditch** – roadside ditches or channel-ways that trap water in low places.
- 6) **lake** – larger than a pond.
- 7) **cattail wetland** – typical of waterbodies that are considered to be true wetlands (i.e., Greenbottom Swamp or Canaan Valley).
- 8) **Other**  
Include comments in the area provided to elaborate on any of these.

## PAGES 5, 6, 5a, and 6a

### EPA's Rapid Habitat Assessment Form

The habitat assessment approach used in this protocol is adapted from EPA's Rapid Bioassessment approach and refined from various applications across the country. The approach focuses on integrating information from specific parameters on the structure of the physical habitat. Specific instruction and training are necessary for an adequate assessment of habitat quality. For each habitat parameter listed, carefully read the description under each ranking category and place the score in the left margin that best describes the condition of the 100 m stream assessment area.

Different assessment forms are used for streams that are riffle/run prevalent versus those that are pool/glide prevalent. After making the initial survey of the stream assessment area, classify the stream as either riffle/run or glide/pool prevalent based on your visual assessment of the dominant habitat type (Note: glide/pool habitats will require "MACS" macroinvertebrate sampling methods for low gradient streams). The WAP sampling strategy dictates that a riffle/run habitat is sampled **if possible**. If a stream reach is mostly glide/pool but has a small area of riffle/run, sample the riffle/run if there is enough to obtain the 1 m<sup>2</sup> of substrate. Accordingly, fill out the **riffle/run** Rapid Habitat assessment form. A glide/pool habitat form should only be used when the MACS sampling method is used. **IMPORTANT: In general, MACS sites are not assessed unless indicated on the stream list or there is a special interest in obtaining data from the site. The MACS technique should only be used in streams that are truly "wetland like", such as sites impounded downstream and offer very little to no observable flow. A general rule of thumb is if you have a difficult time determining which direction the stream is flowing, then MACS methods are probably applicable. MACS methods can also be used on large streams that are too deep to wade. In these larger streams, samples are collected from the bank by jabbing the net into appropriate habitat types. Furthermore, if a stream is heavily embedded with sand but has a perceivable flow, it should not be sampled by MACS methods. Riffle/run protocols should be followed (i.e., benthic samples should be collected by kicking the sand). Also, MACS methods should only be used if there exist enough good habitats to collect all 20 jabs/sweeps.**

NOTE: In low water conditions, many of the RBP parameters will be rated lower than their potential. Do not try to envision a full stream channel (bank to bank) when rating the parameters. Rate the stream conditions as they exist on that day. For example, in low flow conditions the epifaunal substrate/available fish cover parameter would be rated lower than its potential simply because the habitat components are not covered with water during that visit.

## **RIFFLE/RUN PREVALENCE**

1. EPIFAUNAL SUBSTRATE/AVAILABLE FISH COVER: Epifaunal substrates are essentially the amount of niche space or hard substrates (stones, snags) available for insects and snails. Numerous types of insect larvae attach themselves to rocks, logs, branches, or other submerged substrates. The greater the variety and number of available niches or attachments, the greater the variety of macroinvertebrate life will exist in the streams. Rocky bottom areas are critical for maintaining a healthy variety of insects in most high-gradient streams.

Fish cover includes the relative quantity and variety of natural structures in the stream such as fallen trees, logs, and branches, large rocks, and undercut banks, that are available for

refugia, feeding, or laying eggs. A large variety of submerged structures in the stream provide aquatic organisms with a large number of niches, thus increasing the diversity.

2. **EMBEDDEDNESS**: refers to the extent to which rocks (gravel, cobble, and boulders) are covered or sunken into the silt, sand, or mud of the stream bottom. Generally, as rocks become embedded the surface area available to macroinvertebrates and fish (shelter, spawning, and egg incubation) is decreased. To estimate the percent of embeddedness, observe the amount of silt or finer sediments overlying and surrounding the rocks. If kicking does not dislodge the rocks or cobble, they may be greatly embedded. It is useful to observe the extent of dark area on the underside of a few rocks. **To avoid confusion with sediment deposition (habitat parameter number 5), observations of embeddedness should be taken in the upstream and central portions of riffles and cobble substrate areas. Collaborate with the biomorph on this parameter.**

3. **VELOCITY/DEPTH REGIMES**: examines the availability of each of the four primary current/depth combinations: (1) slow-deep, (2) slow-shallow, (3) fast-deep, and (4) fast-shallow. The best streams in high gradient regions will have all four habitat types present. The presence or availability of these four habitats relates to the stream's ability to provide and maintain a stable aquatic environment. The general guidelines are 0.5m depth to separate shallow from deep, and 0.3 meters/second to separate fast from slow.

4. **CHANNEL ALTERATION**: is a measure of large-scale changes in the shape of the stream channel. Many streams in urban and agricultural areas have been straightened, deepened, or diverted into concrete channels often for flood control purposes. Such streams have far fewer natural habitats for fish, macroinvertebrates, and plants than do naturally meandering streams. Channel alteration is present when a stream runs through a concrete channel; when artificial embankments, riprap, and other forms of artificial bank stabilization or structures are present; when the stream is very straight is very straight for significant distances; when dams and bridges are present; and when other such changes have occurred. Scouring is often associated with channel alteration. In some instances, channel alteration may benefit the stream (e.g. K-dams). This parameter should be rated regardless of the intent of the channel alteration. *Note that in the example of K-dams, the channel alteration would be depressed by the presence of these structures, but the Epifaunal Substrate/Available Fish Cover and/or Velocity/Depth Regime score could possibly benefit from their presence.*

5. **SEDIMENT DEPOSITION**: measures the amount of sediment that has accumulated and the changes that have occurred to the stream bottom because of the deposition. Deposition occurs from large-scale movement of sediment caused by watershed erosion. Sediment deposition may cause the formation of islands, point bars (areas of increased deposition usually at the beginning of meanders that increase in size as the channel is diverted toward the outer bank) or shoals, or result in the filling of pools. Increased sedimentation also results in increased deposition. Usually this is evident in areas that are obstructed by natural or man-made debris and areas where the stream flow decreases,

such as bends. High levels of sediment deposition create an unstable and continually changing environment that becomes unsuitable for many organisms.

**To avoid confusion with embeddedness (habitat parameter number 2), observations of sediment deposition should be taken in pools and slow water depositional areas.**

Upstream Watershed Sediment Deposition Total: In addition to rating the 100m reach, if any portion of the upstream watershed was observed, please rate the Sediment Deposition for the section observed. It is not required to drive up the watershed for observations, but rather as a way to take advantage of situations where the upstream area is observed on the way to or from a site.

6. RIFFLE FREQUENCY: is a way to measure the sequence of riffles occurring in a stream. Riffles are a source of high quality habitat and diverse fauna. Therefore, an increased frequency of occurrence greatly enhances the diversity of the community.

7. CHANNEL FLOW STATUS: is the degree to which the channel is filled with water. The flow status will change as the channel enlarges or as flow decreases as a result of dams and other obstructions, diversions for irrigation, or drought. When water does not cover much of the streambed, the amount of useable substrate for aquatic organisms is limited. Do not count extremely large substrate (giant boulders) particles that would rarely if ever be submerged or used by aquatic organisms.

8. BANK STABILITY: measures whether the stream banks are eroded (or have the potential for erosion). Steep banks are more likely to collapse and suffer from erosion than gently sloping banks and are therefore considered unstable. Signs of erosion include crumbling, unvegetated banks, exposed tree roots, and exposed soil. However, exposed cliff faces or rocks provide a stable, non-erodible bank. In addition, the extent to which the bank has healed over with vegetation and roots (*i.e.*, the age of the erosional scars) must be considered. **This parameter is scored by considering right and left banks separately throughout the entire 100 m assessment area.** For example, after observing the right bank it was determined that less than 5% of the total bank area in the 100 m assessment reach exhibited erosional scars. This would result in an optimal score in the range of 9-10.

Upstream Watershed Bank Stability Total: In addition to rating the 100m reach, if any portion of the upstream watershed was observed, please rate the Bank Stability for the section observed. It is not required to drive up the watershed for observations, but rather as a way to take advantage of situations where the upstream area is observed on the way to or from a site.

9. **BANK VEGETATIVE PROTECTION**: measures the amount of the stream bank that is covered by natural vegetation for the area (large trees, small trees, herbaceous layer for most of WV streams). For WAP assessments, the stream bank extends from the edge of the channel floor up to the crest-over at top of bank. The top or "crest-over" of the bank can be determined by looking for an obvious slope break that differentiates the channel from a flat floodplain higher than the channel. The root systems of plants (trees, shrubs, grasses) growing on stream banks helps hold soil in place, thereby reducing the amount of erosion that is likely to occur. This parameter supplies information on the ability of the bank to resist erosion, as well as additional information on the uptake of nutrients of by the plants, the control of in-stream scouring, and stream shading. Consideration must be given to the abundance and diversity of trees, shrubs, or grasses (grazed/mowed and ungrazed/unmowed). The frequency or age of mowing and grazing can also be considered. Banks that have full, diverse, natural plant growth are better for fish and macroinvertebrates than are banks without vegetative protection or those shored up with concrete or riprap. However, the presence of exposed cliff faces or rocks should not detract from this score as they are natural structures that normally do not support vegetation. **This parameter is scored by considering right and left banks separately throughout the entire 100 m assessment area.**

Upstream Watershed Bank Vegetative Protection Total: In addition to rating the 100m reach, if any portion of the upstream watershed was observed, please rate the Bank Vegetative Protection for the section observed. It is not required to drive up the watershed for observations, but rather as a way to take advantage of situations where the upstream area is observed on the way to or from a site.

10. **WIDTH OF UNDISTURBED VEGETATION ZONE**: is a measure of disruptive changes to the natural vegetative zone (big trees, small trees, herbaceous layer for most of WV streams) because of grazing or human interference (e.g. mowing). In areas of high grazing pressure from livestock or where residential and urban development activities disrupt the riparian zone, the growth of a natural plant community is impeded. Residential developments, urban centers, golf courses, and pastureland are the common causes of anthropogenic effects on the riparian zone. **This parameter is scored by considering right and left banks separately throughout the entire 100 m assessment area.**

Upstream Watershed Width of Undisturbed Vegetation Zone Total: In addition to rating the 100m reach, if any portion of the upstream watershed was observed, please rate the Width of Undisturbed Vegetation Zone for the section observed. It is not required to drive up the watershed for observations, but rather as a way to take advantage of situations where the upstream area is observed on the way to or from a site.

**TOTAL**: Total all of the scores for a final RBP score from 0-200.

Estimated Mileage of Upstream Watershed Evaluated: Indicate the approximate mileage of the upstream watershed that was observed for the Upstream Watershed scores.

#### **GLIDE/POOL PREVALENCE FORM (MACS SITES ONLY)**

1. EPIFAUNAL SUBSTRATE/AVAILABLE FISH COVER: See No. 1 under PAGE 5a - RIFFLE/RUN PREVALENCE. In low gradient streams with muddy bottoms, the epifaunal substrate consists mostly of submerged logs or snags, and aquatic vegetation.

2. POOL SUBSTRATE CHARACTERIZATION: evaluates the type and condition of bottom substrates found in pools. Firmer sediment types (e.g., gravel, sand) and rooted aquatic plants support a wider variety of organisms than a pool substrate dominated by mud or bedrock and no plants. In addition, a stream that has a uniform substrate in its pools will support far fewer types of organisms than a stream that has a variety of substrate types.

3. POOL VARIABILITY: rates the overall mixture of pool types found in streams, according to size and depth. The four basic types of pools are large shallow, large-deep, small-shallow, and small-deep. A stream with many pool types will support a wide variety of aquatic species. Rivers with low sinuosity (few bends) and monotonous pool characteristics do not have sufficient quantities and types of habitat to support a diverse aquatic community. As a general guideline: consider a pool deep if it is greater than 1 meter in depth, and large if its length, width, or oblique dimension is greater than half the stream width.

4. CHANNEL ALTERATION: See No. 5 under RIFFLE/RUN PREVALENCE.

5. SEDIMENT DEPOSITION: See No. 6 under RIFFLE/RUN PREVALENCE.

6. CHANNEL SINUOSITY: evaluates the meandering or the relative frequency of bends in the stream. Streams that meander provide a variety of habitats for aquatic organisms, whereas straight stream segments are characterized by monotonous habitats that are prone to flooding. A high degree of sinuosity creates a variety of pools and reduces the energy from surges when the stream flow fluctuates. The absorption of this energy by bends protects the stream from excessive erosion and flooding.

7. CHANNEL FLOW STATUS: determines the percent of the channel that is filled with water. The flow status will change as the channel enlarges or as flow decreases as a result of dams and other obstructions, diversions for irrigation, or drought. The water will not cover as much of the streambed, thus decreasing the amount of living space for



aquatic organisms. In muddy bottom streams, the decrease in water level will expose logs and snags, thus reducing the areas with good habitat.

8. BANK STABILITY: See No. 8 under RIFFLE/RUN PREVALENCE.

9. BANK VEGETATIVE PROTECTION: See No. 9 under RIFFLE/RUN PREVALENCE.

10. WIDTH OF UNDISTURBED VEGETATION ZONE: See No. 10 under RIFFLE/RUN PREVALENCE.

TOTAL: Total all of the scores for a final RBP score from 0-200.

## PAGE 7

### Non-RBP Parameters

BENTHIC MACROINVERTEBRATE SUBSTRATE: This parameter measures the quality of the benthic macroinvertebrate substrate throughout the 100m assessment reach (**this measure excludes the fish component as given in RBP 1. Epifaunal Substrate / Available Fish Cover**). **Only benthic macroinvertebrate substrate quality should be considered with this parameter.** Benthic macroinvertebrate substrate is essentially the amount of niche space or hard substrate (stones, snags) available for insects, snails, worms, clams, and crustaceans to colonize. Numerous types of benthic organisms attach themselves to rocks, logs, branches, or other submerged substrates. The greater the diversity and abundance of available niches for attachment the greater the diversity and abundance of benthic macroinvertebrates in the stream. Rocky bottom areas are critical for maintaining a healthy variety of insects in most high-gradient streams. The relative amount of cobble drives this parameter as it is the most productive substrate size-class for benthic macroinvertebrates in riffle/run samples. The relative amount of transient particles such as fine gravel and sand is also important to consider when rating this parameter.

AESTHETIC RATING: Record the aesthetic character of the stream assessment area (**NOT JUST IN THE STREAM**) based on the abundance of human refuse that is present in and around the stream bank. Consider any piece of trash that could potentially be washed into the stream by high flows or floods. This is also known as the **TRASH INDEX**.

REMOTENESS RATING: Record the remoteness of the stream assessment area based on its proximity to roads and development activities.

### PRS and Stressor Info

**POTENTIAL REFERENCE SITE:** Answer Yes or No. Indicate whether this site appears to be relatively undisturbed and may be considered as a potential reference site (see reference site criteria). Also make notes as to why the stream does not satisfy reference site criteria in the space provided. **Note that a yes answer will not necessarily mean the site will achieve reference status as many other criteria that cannot be determined in the field are considered. Many sites that a person would typically say no to as a potential reference site still meet all of the reference criteria. Therefore it is important to consider only those criteria that can absolutely be determined in the field when answering this question.**

**Stressor Info:** Indicate all definite stressors that are believed to have a impact on the benthic macroinvertebrate community at the site. Options include: Sediment, Fecal and/or Nutrients (both considered Organic Enrichment), Metals (or acid metals which represent toxicity), pH (low pH playing a role in metal toxicity and high pH playing a role in ionic stress), Sulfate and/or Conductivity (both considered ionic stressors), and other stressors.

**Relative Bed Stability (Pebble Count) including Gradient**

This area is provided to record measurements made on the stream substrate and stream channel. Record the measurements in the spaces provided and make comments as necessary. **See SOP L-Substrate Characterization for instructions on completing this section.**

**Reach Length:** Record the total reach length in meters (100m minimum to 500m maximum)

**Measurer:** Record the measurer's initials

**Recorder:** Record the recorder's initials

**Gradient Method:** Check the box corresponding to the gradient method used (water-filled tube or Hand-level)

**Wetted Width:** Record the wetted width in m for that transect

**Left, Left Mid, Middle, Right Mid, and Right:** Record the substrate classification scores for these locations on the transect using the scale in Figure B-4.

**Figure B-4. Substrate Size Classes for Pebble Counts**

Class	Code	Size	Description
Bedrock	BR	>4000 mm	Bigger than car
Boulder	BL	>250-4000 mm	Basketball to car
Cobble	CB	>64-250 mm	Tennisball to basketball
Coarse	CG	>16-64 mm	Marble to Tennisball

Gravel			
Fine Gravel	FG	>2-16 mm	Ladybug to marble
Sand	SA	>0.06-2 mm	Gritty between fingers
Silt & Fines	ST	<0.06 mm	Smooth, not gritty (silt & muck)
Clay	CL	>4000 mm	Slick/ hard clay or hard-pan clay bottom
Leaves	LD	Regardless of size	Leaf packs
Wood	WD	Regardless of size	Rootwads, snags, logs, sticks

Thalweg: Record the thalweg depth in m for that transect

Bankfull Height: Record the bankfull height in m for that transect

Rise: Record the stream rise in m for the distance between transects

## PAGE 8

### Wildlife and Freshwater Mussel Observations

Note actual wildlife or plants observed or indications of their presence (minnows common, kingfisher observed, frog observed, etc.). **Any organisms observed and put into the Benthic Sample Jar should be noted on page 9 under Benthic Sample Notes.**

Common Name: The common name of the organism observed.

Genus/Species: The genus or species of the organism observed.

Comments: Specific notes concerning the organism or evidence of organism observed.

Number Observed: The number of individuals of that organism observed.

Observed: The initials of the observer.

Did you see fresh water mussels?: Answer Yes or No.

Alive or Dead?: Answer Alive or Dead.

Did you collect dead shells?: Answer Yes or No. Dead shells are submitted to Doug Wood for identification or further identification by WVDNR.

## PAGE 9

## Benthic Macroinvertebrate Collection Information

Benthic Sample Collected?: Answer Yes or No.

Why?: Provide reason why benthic sample was not collected.

Bug Device: Indicate which device was used to collect benthic macroinvertebrate samples (bugs). See "Protocols for Collecting Benthic Macroinvertebrates" for a detailed description of each device and its applicability. Describe any deviations from the protocols below. (1) Rectangular frame dip-net, (2) D-net, (3) Hand pick. **Note: Hand-pick methodology is not a comparable method and should only be used if indicated as an alternative on the stream list.**

Habitat Sampled and # of Each: See "Protocols for Collecting Benthic Macroinvertebrates" for a detailed description. (1) **Riffle**, (2) **Run**, (3) **Woody snags (MACS)**, (4) **Vegetated banks (MACS)**, (5) **Aquatic plants (MACS)**.

Benthic Sample Comparability: Was benthic sample comparable with respect to riffle/run depth and velocity? Yes or No: Answer Yes or No. Sampling should generally occur only if the depth is at least 0.05 m deep and has enough velocity to push debris into the net.

Evidence of scouring? Yes or No: Answer Yes or No. Consider asking locals, look at new or recently deposited materials on banks, consider recent precipitation and flood events for the area.

Evidence of dry conditions?: Yes or No: Answer Yes or No. Look for indications that the stream was dry or partially dry recently). Consider asking locals, past weather conditions, benthic macroinvertebrate density and diversity, and stream conditions while you are there.

Evidence of wet-weather stream: ? Yes of No: Consider asking locals, look for dirt channel, vegetation and roots in channel growing across the stream, jagged rocks in the stream, no easily definable U-shaped channel, over abundance of leaves in the stream for the season. Consider watershed area, consider benthic density, diversity, and community composition while collecting sample.

Kick Area Depths (m): Record the measured depth of water at each kick sample location (usually four locations).

A blank space is provided to describe the site and explain responses to the previous questions regarding the sample comparability. Also, any organisms observed *in the sample* should be recorded here.

## Benthic Substrate Sample Composition

Inorganic Substrate Components: Using Figure B-5 as a guide, provide a visual estimate of the relative proportion of each of the seven particle types listed. **This assessment should be conducted only within the actual benthic collection area and should be done by the Biomorph.** In other terms, estimate the proportion of each substrate type within the 1m<sup>2</sup> riffle/run area that was sampled using the following scale:

**Figure B-5. Substrate Size Classes for Pebble Counts**

Class	Code	Size	Description
Bedrock	BR	>4000 mm	Bigger than car
Boulder	BL	>250-4000 mm	Basketball to car
Cobble	CB	>64-250 mm	Tennisball to basketball
Coarse Gravel	CG	>16-64 mm	Marble to Tennisball
Fine Gravel	FG	>2-16 mm	Ladybug to marble
Sand	SA	>0.06-2 mm	Gritty between fingers
Silt & Fines	ST	<0.06 mm	Smooth, not gritty (silt & muck)
Clay	CL	>4000 mm	Slick/ hard clay or hard-pan clay bottom

Low gradient (MACS) streams will require a visual estimate of the entire 100 m assessment area.

Describe Quality of Benthic Substrate: Describe the quality of the benthic macroinvertebrate substrate by noting outstanding features like “nice stacked cobble”, “very sandy and gravelly”, “boulders with a few gravels here and there”, “large amounts of partially broken down leaf packs among the cobble”. Indicate if you think it is stable and capable of maintaining benthic populations.

### Periphyton Collection Information

Periphyton Sample Collected?: Answer Yes or No.

Why?: Provide reason why periphyton sample was not collected.

Periphyton Habitat and #: Record the number of rocks selected from riffles and from runs during periphyton collection,

Shade and #: Record the number of rocks selected from the various shade categories during periphyton collection. Example: 2 in Fully Exposed, 1 in Fully Shaded, and 2 in Partly Shaded.

Periphyton Sample Comparability: Was periphyton sample comparable (consider questions above about benthic comparability)?: Answer Yes or No.

A blank space is provided to describe the site and explain responses to the previous questions regarding the sample comparability.

### **Visual Estimation of Periphyton and Aquatic Plant Density**

Abundance of Periphyton, Filamentous Algae, Aquatic Plants, "Aquatic" Moss: Indicate the abundance of periphyton, algae, aquatic plants, and "aquatic" mosses in the stream assessment area as **none (0), low (1), moderate (2), or high (3)**.

Periphyton is algae, diatoms, fungi, bacteria, protozoa, and associated organic matter associated with stream channel substrates. They are useful indicators of water quality because they respond rapidly and are sensitive to a number of human disturbances, including habitat destruction, contamination by nutrients, metals, herbicides, and acids. In this section of the WAP assessment, periphyton will include only the microalgae. These are the microscopic organisms that make the substrate slick and slimy. They usually leave a brownish-yellow stain on your hand when rubbed.

Although generally included in the broad class of periphyton, filamentous algae (macroalgae) will be considered separately in this section. Filamentous algae include the long stringy types that are green in coloration and exhibit wavy undulations in stream currents. **Note: during Periphyton collection, both the microalgae and Filamentous Algae are collected (see SOP J-Periphyton).**

Aquatic plants are generally associated with larger streams such as the New River and Cacapon River. Riverweed is an example that would be included in the aquatic plant category.

"Aquatic" mosses are those mosses found under the water level in a stream that is typically dry for extended periods and is a good indicator of stream intermittency as well as an excellent benthic macroinvertebrate habitat.

## **PAGE 10**

### **Landowner/Stakeholder Information**

If a landowner or stakeholder encountered during the sampling event expresses interest in obtaining WAP data, record name address and/or phone number here.

Name: Name or names of the landowner or company that owns the land.

Address: Mailing address of the landowner.

Stream Data Requested?: Were the results from this sample requested by the landowner?  
Check Yes or No.

Watershed Report Requested?: Was a future watershed report from the watershed being sampled requested by the landowner? Check Yes or No.

Phone: The primary phone number of the landowner.

Alt #: A secondary phone number of the landowner.

Site Accessibility: A set of check boxes is provided to give a quick indication of what may be involved in getting to the site. Check all that apply. These boxes include: Easy Access, Difficult Access, Private Property, Posted, Fenced, Gated, Get Key from Landowner, Beside Road, Short Hike, Long Hike, 4x4 Needed, or other (explain).

Landowner Notes: A blank field is provided to discuss the accessibility to the site including elaborations on the Site Accessibility check boxes discussed above. This location is also be a good place to keep track of people you talked to while trying to track down the landowner. In the case of a mistake landowner identity, this chain of information will help alleviate any misunderstandings between the field crew and true landowner. Any information about the watershed that may affect sampling should be recorded on Page 2 under the Comment Box describing the source of the information as the landowner.

## **Photography**

A more detailed description of the photography process can be found in **SOP K-Photography**

Camera Type: The type of camera used (e.g., Kodak or Sony Mavica).

Camera Number: The assigned number of the camera used.

Photo ID # (Office): Obtained in the office after getting the unique number from the WAPbase.

Disk-Photo # (Field): A way to keep track of what photo is on what floppy disk and the order of the photos. Each camera assigns these numbers to photos in series from 0-999 or to the capacity of the camera.

Stream Name: The name of the stream featured in the photo. *This is only required if the photo was not taken at a sample site.*

AN-Code: The AN-code (if known) of the stream featured in the photo. *This is only required if the photo was not taken at a sample site.*

Photo Description: A description of the photo as it relates to the stream (e.g., looking upstream from X-site) and the keyword features that may be found in the photo (e.g., AMD, eroded bank, channelization, an optimal score for bank vegetative protection, a poor score for sediment deposition, etc.) This is required for all photos taken.

Date: The date the photo was taken. *This is only required if the photo was not taken on the same date as the sample or if it is not at a sample site.*

Photographer: The person who took the photo. *This is required for all photos taken.*

Sample ID: The designation for that sample that will tie the photo to the other information about that site. *This field is filled out when the data is entered into the database. If a photo was not taken at a sample site, a "0" should be put in this box to help note those photos that are not from that sample site.*

## **APPENDIX 1**

### **Stream Discharge (Flow)**

This area is provided to record measurement made with a flow meter and the resulting CFS (cubic feet per second). Record the Flow Meter I.D., measurer and the time of measurement. Instructions for determining stream discharge (flow) are presented in SOP K- Measuring Stream Flow

Measurer: Record the flow measurer.

Time: The time of the flow measurement.

Flow Meter I.D.: The assigned number of the flow meter used.

Distance: Record distance from one bank along the flow transect (measuring tape) where the measurement is occurring.

Depth: Record the depth at the point of the flow measurement.

Velocity: Record the velocity at the point of the flow measurement.



Total Discharge: Record the total stream discharge by entering in the Distance, Depth, and Velocity data from each increment into the Flow Spreadsheet or from the gage reading.

Is flow measurement comparable?: Answer Yes or No.

If not, why?: Why it is believed the flow measurement is considered not comparable.

USGS Gage Number: The ID number of the USGS gage queried for flow data.

Time: The time the gage was read for the flow measurement.

Gage Height or Control: The Height of the water on the USGS Gage.

## **QUALITY ASSURANCE/QUALITY CONTROL**

Prior to the field season, all participants in the WAS attend a mandatory training session. WAS members will visit one or two stream sites and each person will complete a habitat assessment form at each site. The results of these evaluations will be compared and the group will discuss problems with variability.

The team member who did not conduct the initial assessment performs an on-site review of every habitat assessment. The reviewer determines completeness and verifies that the information is correct through discussion with the other crew member.

Replicate assessments are conducted at 2.5% of the sites. These sites are randomly selected. Every effort is made to assure that different teams perform the replicate sampling. Replication consists of sampling the site by each individual as if no one else was there to help (*i.e.*, one person serves as both Biomorph and Geomorph). Sampling occurs in the usual fashion with the Geomorph doing the habitat assessment and the Biomorph collecting benthos. To replicate, these individuals reverse roles while keeping their data and samples completely separate. The replicate assessments will be examined for comparability. Retraining will be conducted, if major discrepancies are encountered.

## **SOP C. GLOBAL POSITIONING SYSTEMS**

### ***INTRODUCTION***

GPS units use satellite communications to accurately determine the latitude and longitude of a specific location. Since the GPS units use triangulation to determine location, the more satellites it is in contact with, the more accurate the data. To function efficiently the GPS must be used in an unobstructed area and must be in communication with at least four satellites.

### **Garmin III+ or V GPS Unit**

- 1) **Procedures for obtaining coordinates with a GARMAN GPS III+ or V**
  - A) Unfold the antenna.
  - B) Press the red light bulb button to turn unit on.
  - C) At the warning screen, press enter to proceed to the satellite screen.
  - D) Wait an adequate amount of time while the unit locks onto the satellites. The bars at the bottom of the screen will rise with increasing signal strength and will turn black when the signal is locked.
  - E) When the unit has locked into enough satellites to get a reading it will display a map.
  - F) Push the "quit" button twice to get back to the satellite screen. If the reading is adequate, record the EPE. If not wait some more to see if it improves. If it does not, then proceed with what is available or utilize alternative means to determine coordinates (e.g., WCMS, Topo Map, etc.).
  - G) Push the "quit" button until the latitude and longitude are displayed in the lower third of the screen.
  - H) Record the latitude and longitude as "field readings" on the habitat sheet. The Garman units do not store the data nor can the readings be corrected.

## **2) Procedures for checking/changing the datum with a GARMAN GPS III+ or V**

Sometimes it may be necessary to check the datum being used by the unit (e.g., when a unit has been without batteries for an extended amount of time or with the purchase of a new unit). Each datum is different and will dictate how the coordinates be displayed or recorded. Since most of our GIS needs in the office are fulfilled through WCMS, we need to make sure that any data taken or recorded in the same datum used by WCMS. The older 2.8 version of WCMS uses NAD 1927 CONUS for a datum. The newer WCMS version (9.0) will use NAD 1983 CONUS.

- A) Unfold the antenna.
- B) Press the red light bulb button to turn unit on. Wait for the Acquiring Sats screen to appear.
- C) Press Menu twice to get the Main Menu.
- D) Scroll down to Setup and press ENTER.
- E) Scroll right along the tabs to Position.

If "NAD27 CONUS" is displayed under Map Datum, then nothing needs to be changed. Press QUIT twice to get back to the Acquiring Sats screen and turn off the unit.

If "NAD27 CONUS" is not displayed under Map Datum, then scroll down and select whatever is listed under Map Datum. This will cause a list to pop up on the left. Scroll down and select "NAD27 CONUS"; press Enter. The proper datum should now be selected. Press QUIT twice to get back to the Acquiring Sats screen and turn off the unit.

## **QUALITY ASSURANCE/QUALITY CONTROL**

All field personnel are trained in the use of the GPS units during the annual Watershed Assessment Section training session. The instructions listed above are included in the training manual.

The accuracy of the GPS readouts is checked during the data entry phase. Any position that does not fall within the expected location is recalculated using the

Watershed Characterization and Modeling System Arcview GIS program or by a revisit to the site.

## **SOP D. WATER QUALITY SAMPLE COLLECTION, HANDLING AND ANALYSIS**

### ***MATERIALS AND REAGENTS***

1. "Analysis Request Form" - for sample identification and tracking, and maintaining chain-of-custody.
2. Waterproof pen - for labeling sample bottles.
3. Sterile Fecal bottles with sodium thiosulfate tablet - for collecting bacteria samples.
4. Plastic Bottles (Cubitainers with Lids) - for collecting other water quality samples, except phenols.
5. Cooler - for sample preservation.
6. Ice - for sample preservation.
7. Fixatives (nitric acid, sulfuric acid, and sodium hydroxide) - for sample preservation.
8. Waterproof plastic bags or other suitable container - for holding bacteria sample bottles during transport.
9. Filtration Apparatus (either Peristaltic or Vacuum type) – for sample preservation.

### ***SAFETY PRECAUTIONS***

Rubber gloves and protective eyewear should be worn during sample collection and preservation.

Bottles containing fixatives should be stably seated inside a lidded container to prevent breakage.

**WARNING!! SOME FIXATIVES ARE CORROSIVE AND MAY EMIT TOXIC FUMES. BE SURE TO USE THE APPROPRIATE SAFETY GEAR. DO NOT FIX SAMPLES IN THE VEHICLE, AS ACCIDENTAL SPILLS CAN AND WILL OCCUR.**

Do not place liquid acid or base into sample bottles prior to sample collection.

Always add fixatives to sample. **NEVER ADD SAMPLE WATER TO LIQUID ACIDS OR BASES, AS A STRONG CHEMICAL REACTION CAN OCCUR.**

## ***INTRODUCTION***

The water quality monitoring is the centerpiece of the Watershed Assessment Section's efforts to assess streams. It is extremely important that all of these methods are followed to maintain comparability between samplers and sampling events.

## **I. PROCEDURES FOR COLLECTING WATER QUALITY SAMPLES**

1. *Using a permanent ink pen*, fill out an "Analysis Request Form" for each sample site. If the fecal sample will be delivered one laboratory and other samples to another, complete two request forms. The person who actually collected the sample must be the person indicated on the form and the one who signs the chain-of-custody.
2. Label each sample container. The following information must be included: Agency Name (WVDEP/WAP), Stream name, Alpha-numeric code (or station ID), date/time collected, and type of fixative added (if applicable).
3. Collect water samples from mid-stream, mid-depth, at the head or tail of a riffle as follows:
  - A. Fecal Coliform Sample:
    - Use pre-sterilized bottle with sodium thiosulfate tablet. Keep the bottle closed until you are ready to collect the sample.
    - Open bottle and handle carefully to avoid contamination. **DO NOT TOUCH THE INSIDE OF THE LID OR BOTTLE.**
    - Using a quick dipping motion, fill the bottle to the 100 ml mark. **DO NOT RINSE OR REFILL THE BOTTLE.** If the bottle is too full, slowly pour a little out.
    - Place cap tightly on bottle and secure cap lock
    - To further reduce accidental contamination, place sample in a larger bottle or waterproof plastic bag before placing on ice
  - B. Other Water Samples:
    - With the exception of phenols, all remaining water quality samples are collected in plastic Cubitainers.
    - Rinse the Cubitainer twice with stream water
    - Fill the Cubitainer with sample water leaving airspace of about 1% of the

sample volume. **The exception: When collecting a sample to be analyzed for Alkalinity (unfixed sample) or Mercury (Metals Sample) as much air as possible should be expunged from the sample container to avoid contamination.**

- Preserve the sample as indicated on the "Analysis Request Form".
4. With the exception of fecal coliform, all samples should be delivered to the lab within the holding times specified in "Standard Methods for the Examination of Water and Wastewater", 18<sup>th</sup> Edition. The holding times are summarized on the Eagle-Picher "Environmental Sampling Guide". The holding time for fecal coliform sample has been expanded by the WAP from 6 hours to 24 hours<sup>1</sup>. However, **fecal samples collected for the TMDL program may need to comply with the six-hour holding time depending on the specific instructions given for that watershed.**
  5. When the sample is delivered to the laboratory, or picked up by a laboratory representative, complete the chain-of-custody section at the bottom of the "Analysis Request Form". Keep the white copy for your records and give the yellow copy to the lab or delivery person.

## **II. Protocols for Sample Filtration with Peristaltic Pump/Drill Apparatus (Dissolved Metals)**

The components of the filtering apparatus are:

1. Peristaltic Pump mounted on Stabilizing Board
2. Power Drill with Pump Adaptor Bit
3. Tygon Tubing
4. Filters (50 mm cellulose acetate membranes with a 0.45 micron pore size); two varieties: Flat Disc or Cartridge.
5. Two sample containers (one for the stream sample and one to receive the filtered water).

It is important to keep the filtering equipment and area around the equipment clean. Try to handle all parts by the exterior components. Fingerprints and other dirt can contaminate samples. The tubing and filters should be kept in their sealed plastic bags until time of use to reduce exposure to dust and other contaminants.

Ideally, the filtering process would occur at streamside by taking the filtered samples

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<sup>1</sup> The six-hour holding time places severe limitations on the amount of time a crew can spend in the field. Since these samples are not being collected for enforcement purposes, WAP has expanded the holding time to 24 hours.

(e.g., dissolved metals and dissolved orthophosphate) directly from the water column. However, this is dependent upon there being a flat, streamside surface to work on and no precipitation that could short the drill. If filtering cannot occur at streamside, the sample water to be filtered should be collected in a clean container that is rinsed twice with stream water. **This container should be the only one that will be exposed to the Tygon tubing and not reused from site to site. Do not filter from the Total Metals sample container as the insertion of the Tygon tubing may contaminate the sample.**

Procedure:

1. Assemble the filtration unit:
  - Place the drill upside down on stabilizing board and carefully insert the bit into the peristaltic pump.
    - The bit may need to be rotated slightly in order to line up with the receiving shaft and engage fully.
  - Place the unfiltered stream sample container near the pump and remove the cap.
  - Open the pump clamp by lifting the lever.
  - Without directly touching the tubing, open the sealed tubing bag and remove about 8 inches of tubing. Place this end into the unfiltered stream sample container. The rest of the tubing can now be manipulated directly with the hands, but avoid touching the other end of the tubing if at all possible. Thread the tubing through the pump and close clamp.
    - If filtering directly from stream, the tubing can be touched with the hands.
    - Place the stream end of the tubing so that sediment is not being collected from streambed.
  - Attach the filter to one end of tubing
    - Handle filter by edges only, with the pressure valve facing toward the pump and stream sample. The cartridge filters should have an arrow indicating the direction of flow.
    - Make sure not to touch the end of the filter that will be discharging into the dissolved sample container.
2. Flush the filter and tubing briefly with sample water by engaging drill slowly for several seconds.
  - Do not collect the flushed water in the filtered container. Discard elsewhere.
3. Rinse the filtered container:
  - Hold the filter at an angle above the mouth of the receiving (filtered water) container at the point of where the tubing is attached.



- This will allow the user to feel if pressure is building up too quickly in the tube and prevent the tube from explosively detaching from the filter and potentially contaminating the filtered sample.
  - Engage the drill slowly and fill the receiving container with about 50 mL of water.
  - Cap the container, shake vigorously and discard filtrate.
  - Repeat.
4. Filtering the sample:
- Engage the drill and fill the receiving container with at least 200 mL, unless otherwise directed. On one liter cubitainers, this location is near the first character on the long diagonal bar on the side of the cubitainer.
    - Use slow drill speeds (never full throttle) to filter the sample, especially when approaching the desired sample about. This method is supposed to be cleaner, not necessarily faster.
    - If you are close to being done and the pressure is building too fast in the tubing, try using a pulsation with the drill speed. This will often get you to the end without having to change the filter.
5. Changing filters:
- Sometimes it becomes necessary to change the filter while in the middle of processing a water sample. This is usually due to the filter membrane becoming overwhelmed with small particles of silt, which causes the sample to filter extremely slowly. The filter can also become clogged with seemingly clear water due to unseen periphyton. This will also cause the filter to be changed. Should the field personnel feel that it is necessary to change the filter to achieve the minimum amount of sample necessary, then the following steps should be taken:
- Cap the receiving container and remove the clogged filter from the end of the tubing.
  - Replace with a clean filter as before in Step 1 being careful not to touch ends of filter.
  - Flush the new filter as in Step 2 and resume filtering.
- Repeat these steps until a sufficient sample is collected. Record on the lab analysis form how many filters were used. This would give the lab an idea about how high the total suspended solids in the sample should be.
6. Discard and restart the sample if:
- The filter is cracked or split during use.
  - The filter is dislodged from tubing while filtering and the unfiltered water contaminates the filtered sample during use. This would be typical if this happens explosively.
  - Sediment is collected directly from bottom of stream.

*NOTE: The tubing and filters are disposable, and should only be used once. Discard the each filter after one use and discard the tubing after each sample. Obtain a clean set for the next sampling event.*

### **III. Protocols for Sample Filtration using a Vacuum Pump (Dissolved Metals)**

The components of the filtering apparatus are:

1. Filter Flask – Receptacle for the filtered sample
2. Filter Funnel – Consists of two parts: A cup to hold the unfiltered sample and the funnel itself.
3. Filters – Cellulose Nitrate membranes with a 0.45 micron pore size.
4. Vacuum Pump – A variety of hand operated pumps are available.

It is important to keep the filtering apparatus clean. Try to handle all parts by the exterior components or by the stopper. Fingerprints and other dirt can contaminate samples. The Filter Funnel & Filter Flask should be stored in a Zip Loc bag or other container (even when driving from one site to another) to reduce exposure to dust and other contaminants.

#### **Procedure:**

The water for the filtered sample must be taken from a portion of the total metals sample.

1. Rinse off the filter apparatus (cup, funnel and flask) with deionized water.
  - Be careful not to get water into the nipple on the flask.
  - Rinse each part separately. Do let rinse water from cup drip into either the funnel or flask and do not let rinse water from the funnel drip into the flask.
2. Assemble the filtration unit:
  - Attach the funnel to the flask.
  - Place a filter on the funnel.
    - Handle the filter by the edges only.
    - Make sure the filter is centered on the funnel's screen.
  - Attach cup, be sure to get a good seal.
3. Initial Rinse:
  - Pour a small amount of sample into cup.
  - Filter sample, making sure all the water has passed through.
  - Depressurize the pump.
  - Wipe drips from exterior of cup & funnel and remove from flask without

disassembling cup from funnel.

- Rinse the flask with a swirling motion and discard filtrate (be careful to avoid getting filtrate in the flask nipple).

#### 4. Filtering the sample:

- Place cup & funnel assembly back into flask.
- Pour a larger amount of the sample into the cup.
  - If water is turbid, use small amounts; filter may clog and need to be changed.
  - Do not put too much sample into the cup since this may exceed the capacity of the flask, causing water to be sucked into the pump.
  - Wipe off any spills outside of the cup.
- Filter sample using full strokes on the pump.
- Depressurize pump after sample has been filtered and before changing filters.

#### 5. Changing filters:

Sometimes it become necessary to change the filter while in the middle of processing a water sample. This is usually due to the filter membrane becoming overwhelmed with small particles of silt which causes the sample to filter extremely slow. Should the field personnel feel that it is necessary to change the filter to achieve the minimum amount of sample necessary, then the following steps should be taken:

- If there is any left, pour off the excess water out of the cup by turning the filter apparatus on its side with the siphon arm up so that no filtered water can escape from the flask or enter the vacuum tube. One should support both the cup and the lower funnel so that the two do not break the magnetic seal and separate.
- Filter off any excess water until the filter is dry.
- Remove the cup from the funnel.
- Holding funnel sideways, remove old filter. Start from the top of the filter and pull downward.
- If there is any question that unfiltered water may have dripped onto the funnel or into the flask, assume that the sample has been contaminated and the filtering process must be reinitiated from the beginning.
- Install a fresh filter handling only by edges.
- Replace the cup and continue filtering.
- Repeat these steps until sufficient sample (usually a net of 200ml of sample after rinsing the cubitainer 1-2 times, but check with lab beforehand). It is also a good idea to put on the lab analysis form how many filters were used if greater than 1. This would give the lab an idea about how high the Total Suspended Solids in the sample should be.

6. Discard sample if:
  - Filter is cracked or split during use.
  - Sediment on filter is off-center (no white ring around entire edge).
  
7. End of week cleaning:
  - Rinse cup, funnel and flask with tap water; wipe off scum.
  - Use a brush to lightly clean the funnel's screen.
  - Rinse cup, funnel and flask thoroughly with deionized water and shake off excess droplets.
  - Place a filter on the funnel's screen and store cup/funnel assembled in a zip loc bag.
  - Rinse only the glass flask with 10% HCl. The plastic portions (funnel and cup) may only be rinsed with deionized water and lightly rubbed with a paper towel.
  - Do not touch inside surfaces of filtration apparatus.

**Note:** When preserving the total and dissolved metals from a vacuum pump filtered sample, use only 1 ampule of HNO<sub>3</sub> split proportionally among the two cubitainers according to the volume of each sample.

#### **IV. Water Quality Parameters**

***Take Hydrolab readings & Fecal coliform at every site!***

##### **Random & Potential Reference Sites:**

- Acidity (Hot), Alkalinity, Sulfate, Chloride, Fecal coli., TSS, Tot. Phos., TKN, NO<sub>2</sub>-NO<sub>3</sub>-N, Mg, Al (Tot. & Dis.), Cu (Dis.), Fe (Tot. & Dis.), Mn, Hg (0.1 ug/L MDL), Zn (Dis.), Ca, Se (Tot. & Dis.). (Note: Order Low Level Detection on Tot. & Dis. Cu, Zn, & Se.)

**4 cubies (iced, HNO<sub>3</sub>, filtered HNO<sub>3</sub>, & H<sub>2</sub>SO<sub>4</sub>) & fecal**

##### **AMD Parameters:**

*Take when: 1) conductivity alone is >500, 2) pH <6.0 & conductivity is >200, 3) if stream is on the 303(d) list for AMD, or 4) if for any reason you suspect mine drainage:*

- Acidity (Hot), Alkalinity, Sulfate, Chloride, Fecal coli., TSS, Al (Tot. & Dis.), Fe (Tot Only), & Mn. Take Ammonia-N (NH<sub>3</sub>) if it is suspected that Ammonia is being used to treat the stream water.

**3 cubies (iced, HNO<sub>3</sub>, & filtered HNO<sub>3</sub>) & fecal**

##### **Acid Rain Parameters:**

*Take when: 1) pH <6.0 & conductivity is <50, 2) if stream is on the 303(d) list for pH unrelated to mining, or 2) if for any reason you suspect acid rain deposition*

*impacting the stream:*

- Acidity (Hot), Alkalinity, Sulfate, Fecal coli., Acidity (Cold), TSS, Al (Tot. & Dis.), Fe (Tot Only), Mn, & Ca (Tot.).

**3 cubies (iced, HNO<sub>3</sub>, & filtered HNO<sub>3</sub>) & fecal**

**Nutrient Enrichment:**

*Take within 24 hours of a significant rain or when animal waste, straight pipes, STP outfalls, etc., may be impacting the stream:*

- TSS, Tot. Phos., TKN, NO<sub>2</sub>-NO<sub>3</sub>-N, & Fecal coli. Take Ammonia-N (NH<sub>3</sub>) if cattle or other livestock have direct access to stream or if there is evidence of possible ammonia input.

**2 cubies (iced, H<sub>2</sub>SO<sub>4</sub>) & fecal**

**Oil & Gas:**

*Take if oil or gas activities are evident & cond. >200 in absence of other sources like AMD:*

- Chloride & Fecal coli.

**1 cubie (iced) & fecal**

**Other Water Quality Notes:**

- Place fecal bottles in separate zip-lock sandwich size baggies before putting in ice (do not submerge in ice water!).
- Label each container w/ WV DEP WAP, stream name, AN-code, date/time collected, collector (especially if a duplicate), & preservative types.
- Take water samples at lower end of reach for Non- Random targeted sites. Take water samples at lower end of 100 m assessment reach for random sites regardless of the location of the X-site.
- If Alkalinity is being analyzed, 100% of the air must be expunged from the unfixed cubitainer to avoid contamination.
- Remember: A net minimum of 200 mL of filtered sample should be turned in for dissolved metal analysis at most labs we deal with.

**QUALITY ASSURANCE/QUALITY CONTROL**

General

The Watershed Assessment Section conducts annual mandatory training sessions to assure that all members are familiar with sampling protocols. These sessions occur in April or May, prior to the initiation of the sampling season. A hands-on session concerning the collection and handling of water quality samples is included.

This document is also included in the training manual that is given to all

participants.

#### Field Blanks (also see SOP E-Field Blanks and Duplicates)

To evaluate sample containers for contamination, each team will prepare field blanks weekly. For Watershed Assessment studies field blank preparation is an assigned task; performed in conjunction with a randomly-selected field evaluation. TMDL field blanks are prepared weekly but not at an assigned location. At a minimum, each team will prepare one field blank for each watershed. Distilled, deionized water is used as the blank "sample". This water should be carried in a well-sealed container. During the designated sampling event, an extra set of Cubitainers are prepared as field blanks, one container for each type of acidic fixative. The blanks are labeled according to the protocols. These containers are filled with the distilled/deionized water and are preserved and stored in the same manner as the actual samples. A separate "Analysis Request Form" is completed for the field blanks and the samples are submitted to the laboratory.

Lot numbers of preservatives are recorded on the "Analysis Request Form" for each sample submitted.

#### Duplicate Sampling (also see SOP E-Field Blanks and Duplicates)

Replicate water quality samples are collected at 2.5% of the sample locations. For Watershed Assessment, the stations to be subjected to replicate sampling are selected randomly via computer program. TMDL replicates are collected at any TMDL AMD/Fecal site. TMDL replicate sites are not specifically assigned; however, field crews should not repeatedly duplicate the same site. Results of the replicates are compared and any samples not falling within an acceptable range are examined for sampling error.

**Note: If two people are involved in collecting a duplicate, each person should filter his or her own sample and not filter the other person's sample.**

## SOP E. FIELD BLANKS AND DUPLICATES

### ***INTRODUCTION***

Duplicate sampling and field blanks must be performed at a minimum of 2.5% of our sites. To assure we meet these requirements, each team list will have a designated duplicate and field blank. Procedures for performing duplicates and field blanks are presented below.

### **I. Field blanks**

Field blanks are simply samples of deionized water that are preserved in the field. The purpose of the field blank is to detect onsite contamination and verify the purity of the sample fixatives.

Before leaving the office, obtain the deionized water by collecting it directly from the water purification system. The still and filtration system are located in Room 10-A. Procedures for obtaining water are as follows:

1. Feel the storage tank with your hand. If the tank is hot, the still has been in operation recently. Do not continue if the tank is warm (>40 degrees C), as this temperature will damage the filtration system. If the tank is cool, proceed to step 2.
2. Turn on the filtration system and allow the pump to run until the ohm meter on the pump reads 18.
3. While the pump is still running, open the valve exiting the filtration system and the valve at the end of the plastic hose and drain the deionized water into a suitable container for five minutes.
4. Discard the water you have just collected, as it may contain contaminants and other residuals that were in the filtration cartridges.
5. Fill up the appropriate amount of cubitainers to be used as field blanks in the future (1 cubitainer per type of preservative).

Field blanks are to be prepared on site at the designated location. If you miss the exact location indicated on the sheet, prepare a field blank at the time you remember. The reason why field blanks are indicated on your list is to remind you to do it AND to assure that field blanks are prepared at random locations and times.

A field blank will consist of any nitric and sulfuric analyses you are collecting that week (or plan to collect). Do not prepare a field blank for fecals, as the deionized water is not sterile.

To prepare a field blank, retrieve your pre-filled cubitainers with DI water from storage. Label them in a manner that it will appear to be an actual water sample to the lab, but will be recognizable as a blank to Watershed Assessment employees. Fix the samples and handle the sample as you would do for a stream sample. This includes filtering for dissolved metals if that was done during the week. After the sample has been submitted to the lab. Write "FIELD BLANK" at the top of the DEP copy (white) of the Analysis Request Form before submitting it with the other forms.

## **II. Duplicate Samples**

### 1. WAP Sites

With the exception of GPS, a WAP site is to be duplicated in its entirety. Each team member should treat the site as though he/she is sampling alone: Do your own habitat, water quality, and benthic and periphyton collection. The two sets of forms and benthic and water samples should be clearly marked with Dup #1 and Dup #2. On-site water quality data (*i.e.*, pH, Conductivity, DO, Temperature) should only be recorded on the first duplicate form (Dup #1).

Sites to be duplicated are indicated on the Team Lists. These sites are randomly selected and the main purpose of indicating these sites is to remind you to perform duplicate sampling and to assure that duplicates are performed at random locations and times. It is possible that the site selected is unsuitable for benthic sampling or has insufficient habitat to conduct duplicate benthos collections. If this is the case, the duplicate can be performed at an alternate site. Additionally, if you encounter a site that is ideal for duplicate sampling before you get to your designated duplicate site, you may conduct the duplicate at that site and drop the designated duplicate. The important thing is that duplicate sampling is performed for the give group of samples.

### 2. TMDL Water Quality

Duplicate Samples for non-aquatic life TMDL samples are limited to water quality only. There is no need to submit a duplicate TMDL-Initial or TMDL-Secondary habitat form, as most field personnel will be working solo.

Duplication will be limited to the water quality parameters assigned to that site; *i.e.*, if the site is fecal only, just do fecal.

Duplicates for TMDL samples should be conducted at sites where the most parameters on the list are collected (if such sites exist on the list) and should be rotated to different sites each sampling event.



## **QUALITY ASSURANCE/QUALITY CONTROL**

The Watershed Assessment Section conducts annual mandatory training sessions to assure that all members are familiar with sampling protocols. These sessions occur in April or May, prior to the initiation of the sampling season. A hands-on session concerning the collection and handling of water quality samples is included. This document is also included in the training manual that is given to all participants.

The field blank and duplicate data are looked at by Watershed Assessment Section staff and scrutinized to find any possible discrepancies, contamination, or faults in the sampling methods and techniques. Any problems are brought to the attention of the program management and steps are made to immediately correct the problem. Data that is related to the problem are flagged with notes concerning the details of the situation so that decisions can be made whether or not to include the data in any further assessments or analysis.

## SOP F. PHOTOGRAPHY

### **INTRODUCTION**

The Watershed Branch needs quality photographs from every site to use as illustrations for our reports, presentations, and for general use. They are vital for illustration and clarification of the ideas presented as well as visual relief from all the words in the text. To achieve this we need the field personnel to take a variety of pictures while they are in the field. Along with the pictures we need a way to keep track of these photos on our field forms as well as in our database.

This "photography log" is essential for four reasons:

1. We need to know who took the picture
2. We need to know where the picture was taken
3. We need to know what the picture is of
4. We need to know what to call the photo

For information about how to take a photograph with a particular camera, use various features, and download the photos to a computer, consult the operation manual with the camera.

### **I. Procedures for In the Field**

Don't hesitate to take more than one picture of the same scene or activity. Vary the picture by using the settings feature on the camera (e.g., flash level, aperture speed, exposure, wide angle/telephoto, etc.). Always use the highest image size setting on the camera. This will take up more space, but it will provide us with the most useable pictures.

Obviously all pictures will not be used in the report for the watershed where they were taken. Or any other report for that matter. But they may be used later in a presentation, brochure, or report we haven't thought of yet. In addition, these photos may be valuable for the 303(d) narrative criteria listings, 303(b) assessments, or TMDL process (e.g., clarify and extent of hydroxides in stream). We cannot have too many pictures to choose from.

We need pictures of such items as:

- ◆ Stream alteration or management practices
- ◆ Stream disturbances
- ◆ Waterfowl or other wildlife in or near streams
- ◆ Silt laden streams flowing into clear streams
- ◆ Scenic Views
- ◆ Field crews at work

- ◆ Distinctive views of streams, buildings along streams, industry along streams, dams, boats or barges or other water related pictures.
- ◆ Pollution sources and features (e.g., point and non-point sources, metal hydroxides, poorly constructed roads, feedlots, etc.)

All pertinent information about a photo should be recorded on the field sheet under the photography log section. This information includes:

**Camera Type:** The type of camera used (e.g., Kodak or Sony Mavica).

**Camera Number:** The assigned number of the camera used.

**Disk-Photo #:** A way to keep track of what photo is on what floppy disk and the order of the photos.

**Stream Name:** The name of the stream featured in the photo. ***This is only required if the photo was not taken at a sample site.***

**AN-Code:** The AN-code (if known) of the stream featured in the photo. ***This is only required if the photo was not taken at a sample site.***

**Photo Description:** A description of the photo as it relates to the stream (e.g., looking upstream from X-site) and the features that may be found in the photo (e.g., AMD, eroded bank, channelization, a optimal score for bank vegetative protection, a poor score for sediment deposition, etc.). **This is required for all photos taken.**

**Date:** The date the photo was taken. ***This is only required if the photo was not taken on the same date as the sample or if it is not at a sample site.***

**Photographer:** The person who took the photo. **This is required for all photos taken.**

**Sample ID:** The ultimate designation for that sample. *This field is filled out when the data is entered into the database. If a photo was not taken at a sample site, a "0" should be put in this box to help note those photos that are not from that sample site.*

## **II. Procedures for In the Office**

### Tagging the Photos with a Photo ID

In order to keep track of so many photos, at the end of each sampling week each team will need to tag each photo with a unique photo ID number that is maintained in the database. The following are the steps required for to not only tag each photo with this photo ID, but also ensure that each photo ID will have a description in the database as well.

#### *1. Photos that are taken at sampling sites*

Most of the photos that we take are of this type and require the least amount of time to prepare for the database.

- A. Open the WAPbase.
- B. Select the Form called "Photo ID Form".
- C. In the top or bottom tool bars, press the "new record" button (It looks like a triangle pointing to the right followed by a star).
- D. Check the box called "Used Number". Once this button is pressed, a number will appear in the box called Assigned Photo ID. These are the only two fields that should have data in them.
- E. Rename the photo using this number as the name (e.g., 136.jpg, 456.jpg, etc.)
- F. On your field sheet, write this number under Photo ID on the line where your photo information is recorded.
- G. Go to step 3 and repeat for more photos or close the database if done.
- H. Copy/Cut/or Move all of the photos from your computer onto the network server at the following directory:  
G:\Wap\WASP\Photos\Coded Photos
- I. If a message appears asking if you want to replace a file, press no. If this happens, then someone has already named a photo by that name and the two photo names (yours and the one already on the server) need to be investigated and resolved.

All of the information on your field sheet will be entered in during the data entry process and can be linked to your photo by the photo ID. The data entry person will write the appropriate sample ID next to each photo taken at that site.

## 2. *Photos that are taken at sampling sites*

Only a handful of photos that we take are of this type. Since they will not be tied into a Sample ID all data entry for these photos is the responsibility of those who took the pictures.

- A. Open the WAPbase.
- B. Select the Form called "Photo ID Form"
- C. In the top or bottom tool bars, press the "new record" button (It looks like a triangle pointing to the right followed by a star).
- D. Check the box called "Used Number". Once this button is pressed, a number will appear in the box called Assigned Photo ID.
- E. Rename the photo using this number as the name (e.g., 136.jpg, 456.jpg, etc.).
- F. Reenter the photo ID number in the box called "Photo ID".
- G. Begin entering the data in the boxes below the Photo ID (*i.e.*, Photo Description, Photographer, Camera Type, Camera Number).
- H. Enter the applicable site information in the green box (*i.e.*, Stream Name, AN-code, Mile Point, Descriptor, Date, Watershed, Latitude and Longitude).
- I. In the bottom of the green box, enter a 0 for the Sample ID only. **Do not enter any information in the red box.**
- J. Go to step 3 and repeat for more photos or close the database if done.

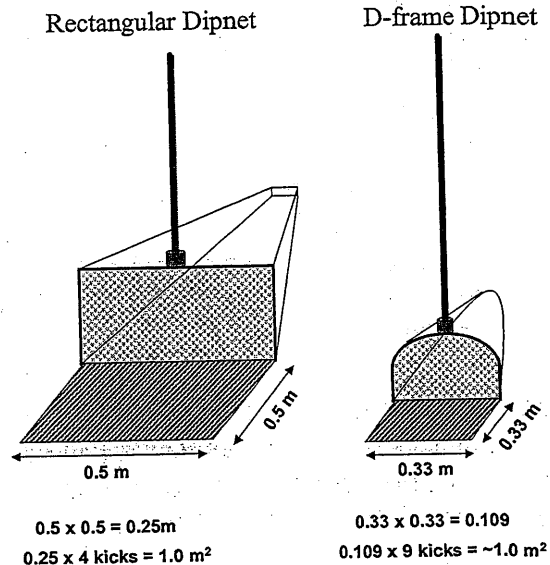
- K. Copy/Cut/or Move all of the photos from your computer onto the network server at the following directory:  
G:\Wap\WASP\Photos\Coded Photos
- L. If a message appears asking if you want to replace a file, press no. If this happens, then someone has already named a photo by that name and the two photo names (yours and the one already on the server) need to be investigated and resolved.

Again, because these photos are not taken at a site, they will not be entered during the data entry process and assigned a Sample ID. The only way information will be entered about these sites is if the crew who took them enters the data. And a photo without this information is not very useful.

## SOP G. BENTHIC MACROINVERTEBRATE COLLECTION PROTOCOLS

### MATERIALS AND REAGENTS

1. Rectangular Frame Dipnet – A net with a 0.5 m wide and 0.3 m high frame with 500  $\mu\text{m}$  mesh openings and 0.5 m nylon bag attached to a four foot pole will be used to collect macroinvertebrates in riffles and runs.
2. D-Frame Dipnet - A D-frame (D-net) aquatic dipnet with 500  $\mu\text{m}$  mesh openings and 1 ft. nylon bag will be used to sample streams that are too small to be sampled using the rectangular dipnet. The D-net will also be used to sample slow flowing streams that do not have riffle/run habitat.
3. Five-gallon Bucket - to composite samples in the field.
4. 30 mesh sieve (600  $\mu\text{m}$ ) - to remove small particulates and water from samples both in the field and in the lab.
5. Small dish washing scrub brush – to aid in removing macroinvertebrates from removable substrate.
6. Sample jars  $\frac{1}{2}$  filled with Denatured Ethanol- containers to hold benthic sample and associated debris.
7. Labels - for sample identification and tracking.
8. 95% Denatured Ethanol - for "fixing" and preservation of benthic macroinvertebrates.
9. Cooler or box - for the storage of samples during transport.
10. Fine Tip Forceps - for removing organisms from net or sieve.
11. Sample Log Book - for tracking the locations of the biological samples.



### SAFETY PRECAUTIONS

Rubber gloves and protective eyewear should be worn during sample collection to avoid bacterial contamination.

## **INTRODUCTION**

### **Benthic Macroinvertebrates as Environmental Indicators**

Benthic macroinvertebrates are small animals living among the sediments and stones on the bottom of streams, rivers, and lakes. Insects comprise the largest diversity of these animals and include mayflies, stoneflies, caddisflies, beetles, midges, crane flies, dragonflies, and others. A benthic community can also have snails, clams, aquatic worms, and crayfish. These animals are extremely important in the food chain of aquatic environments. They are important players in the processing and cycling of nutrients, and are major food sources for fish and other aquatic animals.

Benthic macroinvertebrates have been used for many years to assess water quality. Currently, macroinvertebrates are utilized throughout the world in water quality assessment, as environmental indicators of biological integrity, to describe water quality conditions or health of the aquatic ecosystem, and to identify causes of impairment. Benthic communities are known to respond to a wide array of environmental stressors in different ways. This response will often make it possible to determine the type of stress that has affected the community. Many macroinvertebrates have relatively long life cycles. Thus, community structure is a function of past water quality conditions.

### **Basis of Sampling Method**

The sampling methods to be used in the WVDEP Watershed Assessment Section (WAS) are qualitative in nature and are outlined in "Rapid Bioassessment Protocols for Use in Wadeable Rivers and Streams, Second Edition" - U.S. Environmental Protection Agency, July 1999 (EPA 841-B-99-002). This protocol has been adopted for use by many states. The WAP will utilize the Single Habitat Approach when possible, using a rectangular dipnet (0.5 m wide) or smaller (0.3 m wide) D-net with 500 $\mu$ m mesh size to sample riffle/run habitats. The Multi-habitat Approach will be used in slow-moving wetland type streams, using the smaller D-net. *It is important to note that the following protocols were established for use by the Watershed Assessment Section monitoring program and were intended to provide cost-effective techniques with comparable data across the state. Special projects outside of the Watershed Assessment Section monitoring agenda (i.e., point source surveys, spills, large river monitoring) may not allow strict adherence to these protocols.*

The sampling protocols are listed and prioritized below:

1. Rectangular Dipnet - for riffle habitats  $\geq$  0.5 meter wide
2. D-Frame Net - for riffle habitats  $<$  0.5 meter wide
3. D-Frame Net - used in the absence of moving water (for use in low-gradient streams and glide/pool habitat – Multi-habitat Approach)
4. Hand Picking - used in very small streams where other sampling apparatus cannot be used.

These methods are described in detail in the subsequent sections.

### Selecting Sampling Sites

Predominantly, streams in West Virginia are high gradient with coarse substrate materials such as boulder, cobble, and gravel. These physical conditions are responsible for the typical riffle/run habitats commonly found in most areas of the state. WAP establishes assessment reaches on streams based on the availability of this riffle/run habitat (probabilistic sites excluded). There should be at least one square meter of riffle/run habitat in the assessment reach to obtain a complete benthic macroinvertebrate sample. It is important that the sampling method be selected based on the availability of the reference condition (riffle/run predominant for most of WV) and not of potentially impaired streams. For example, sampling decisions should not be altered for situations where the amount of cobble/gravel substrate in streams influenced by heavy sediment deposition may be substantially reduced from the amount of cobble/gravel substrate expected for the region. That is, sample sites on streams with heavy deposits of fine sediments should not be avoided if it is determined that the sedimentation is not typical of the area and has resulted from poor land-use practices. Occasionally, low gradient streams are encountered that have heavy deposits of fine sediments as a result of naturally high sedimentation rates. In this case, the Multi-habitat Approach should be employed. Currently, WAP does not conduct benthic assessments on low gradient streams unless there is a special interest for the resultant data. The decision to sample a particular stream site is field based and should be made after corroboration by WAP team members or by the most experienced person. In any event, detailed notes describing the situation should be recorded on the field form.

After the 100-meter assessment area has been established, the benthic macroinvertebrate collector (Biomorph) should select sampling points with the intent to make collections throughout the entire 100 meters. In some instances, riffles may be limited to a small area within the reach. In this case, collections should be made within the riffle areas only. However, if riffle areas occur throughout the 100-meter reach, an effort should be made to collect from as many different points within the reach as possible. It is important to sample a diversity of riffle/run conditions if they exist. For example, look for varying conditions within the reach such as fast and slow riffle/runs,



deep and shallow riffle/runs, shaded and exposed riffle/runs, and sample from each observed.

The various habitat types that may be encountered are defined as follows:

**Pool** - Still water with low velocity. Water surface is smooth and glassy. Usually deep compared to other parts of the channel.

**Glide** - Slow moving water with a smooth, unbroken surface. Turbulence is low. Usually shallow compared to other parts of the channel.

**Run** - Similar to glide but water is moving slightly faster. Turbulence is low and the surface is without ripples that produce gurgling sounds. May have small waves.

**Riffle** - Water moving with small ripples, waves and eddies. Produced a babbling or gurgling sound.

**Snag** - Submerged woody debris (logs, root wads, etc.).

**Submerged Macrophytes** - Aquatic vegetation growing beneath the water surface.

**Vegetated and Undercut Banks** - Stream banks having submerged vegetation (shrubs, etc.) and/or root wads.

## ***SAMPLE COLLECTION METHODS***

*Before any sampling event:*

- Fill out a pre-printed sample label with a No. 2 pencil. Attach to the outside of the sample jar using clear, waterproof tape. Fill out a pre-printed sample label made of waterproof paper for the inside of the sample jar.
- Fill the sample jar ½ full with alcohol.
- If using a net, check the net to ensure there are no holes or remnants of previous samples. If there are holes or tears in the net, it should be repaired or replaced as soon as possible and before the next sample is collected.

### **I. Rectangular Dipnet (Riffle/Run Habitats = Comparable)**

This method is used in streams having riffle/run habitat and a width  $\geq 0.5$  meter.

This method is to be used even when there is no cobble substrate in the riffle/run area. If the stream has enough flow to move benthic macroinvertebrates into a net, then this is the method to use.

1. Select a riffle/run area to sample. Position the sampler on the stream bottom so as to eliminate gaps under the frame with the net opening upstream. Large rocks or logs that prevent the net from seating properly should be avoided.
2. Hold the sampler in position on the substrate while checking for heavy organisms, such as clams and snails in an area of about 0.25 m<sup>2</sup> (0.5m wide net x 0.5m upstream) in front of the net. Hand pick these organisms and place them in the net.
3. Brush the surfaces of all cobble, boulder, and bedrock substrate (larger chunks of gravel may also need to be brushed as well). If the substrate is removable, pull it up and hold it in front of the center of the net while brushing all surfaces so that dislodged organisms flow into the net. Cleaned substrate should then be set aside. In low flow situations, these rocks can be placed at the edge of the net in a manner that increases the amount of water flowing through the net. Any large substrate that is only partially in the kick area should only be brushed on that portion which resided in the original 0.25 m<sup>2</sup> kick area.
4. Hold the net handle securely while kicking the substrate vigorously for 20 seconds in an area of about 0.25 m<sup>2</sup> (0.5m wide net x 0.5m upstream) in front of the net. At this time it may be possible to remove large objects (e.g., cobble, large gravel) from the net while the water is still sweeping through the net. Remove the net from the water with a quick upstream motion to wash the organisms to the bottom of the net. Empty the contents of the net into a bucket that is partially filled with water. It is not necessary to remove every last item from the net at this point. Fish, salamanders, and crayfish tend to escape if debris clogs the mesh and subsequently slows the flow of water through the net.
5. Repeat this process until 4 riffle/run habitats have been sampled. This will result in 4 individual kick samples that cover approximately 1 m<sup>2</sup> (4 x 0.25 m<sup>2</sup>) of stream substrate. The 4 kick samples will be composited into 1 sample. If a diversity (fast and slow – stacked and flat, etc.) of riffle/run types is not present, collect the 4 samples from the best available habitat. It is important to obtain 4 kick samples for the composite. Always record the type and number of each riffle/run sampled on the field assessment form. *The RBP protocol (EPA 841-B-99-002) suggests that 2 square meters of substrate*

*should be sampled and composited at a given site. WAP determined through analysis of duplicate data (2 m<sup>2</sup> versus 1 m<sup>2</sup>) and consultation with EPA Region III biologists that a 1 square meter sample is adequate for characterizing riffle/run streams in West Virginia where the West Virginia Stream Condition Index is to be used for impairment classification.*

6. Inspect the net for clinging organisms. Using a pair of small forceps, remove all the remaining organisms and place them in the bucket.
7. While the sample is in the bucket, all large objects (rocks, sticks, leaves, etc.) should be carefully washed, inspected for organisms, and discarded. It is very important to remove as much rough material as possible without losing organisms. This will reduce the sample processing (bug-picking) time substantially. It will also limit the crushing and grinding effect that damages benthic specimens.
8. Elutriate the bucket's organic material (bugs, leaves, CPOM) by using stirring or swirling motion. Begin pouring the elutriated organic material into U.S. Standard 30 sieve. Transfer this material from the sieve into a temporary container (e.g., another bucket, a tray, another sample jar). If possible, release any fish and/or salamanders and document the species and number released in the Wildlife Observations section of the Habitat Form. Repeat this process until almost all of the organic material is gone from the bucket.
9. Begin the elutriation process again with the inorganic material (gravel, sand, silt). Pour the entire contents of the bucket through a U.S. Standard 30 sieve. Using a squirt bottle, rinse any remaining organisms from the bucket onto the sieve. If possible, release all fish and salamanders and document the species and number released in the Wildlife Observations section of the Habitat Form.
10. Using a quiet area of the stream or fresh water in the bucket, gently touch the bottom of the sieve to the water surface and rotate it in a circular motion. This will aid in removing fine sediments from the sample. **DO NOT IMMERSE THE SIEVE ENTIRELY AS THIS WILL RESULT IN THE LOSS OF ORGANISMS.**
11. Pour the inorganic material (gravel, sand, silt) of the sieve into a sample jar already 1/2 filled with alcohol. Use a squirt bottle to aid in removing remnants of the sample from the sieve, but avoid getting large amounts of water in the sample jar, as this will dilute the preservative. Inspect the sieve carefully for any remaining organisms and place them in the sample jar. If you need to drain some water from the sample jar, pour it through the sieve. Then,

recollect any organisms that may have been transferred during the process.

12. Return to the organic material (bugs, leaves, CPOM) that was set aside earlier. Using the same procedure as in Step 9, gently rinse the organic material in the sieve using water surface. Once all of the fine sediments are thoroughly removed, pour all of the organic contents of the sieve on top of the inorganic material (gravel, sand, silt) already in the sample jar as in Step 10. **THESE EXTRA STEPS WILL HELP KEEP FRAGILE MACROINVERTEBRATES FROM BEING DAMAGED BY THE WEIGHT AND ABRASIVE NATURE OF THE INORGANIC SAMPLE MATERIAL. DO NOT AT ANY TIME SHAKE OR INVERT THE SAMPLE JAR ONCE THE ORGANIC MATERIAL IS PLACED INSIDE.**

**When hiking long distances to a sample site and you do not want to carry any extra alcohol in collection jars, follow this procedure.** Take two lidded bug jars to the site: one for the inorganic (sand/silt) portion and one for the organic (bugs/leaves) portion. Upon return to the vehicle, follow the procedures starting with Step 11.

## **II. D-net (Riffle/Run Habitat = Comparable)**

In some situations the stream may be too narrow or shallow to sample using a Rectangular Dipnet. In this case, a D-net will be substituted for sample collection. The methods outlined for the Rectangular Dipnet are applicable when using the D-net in riffle/run streams. The only modification is an increase in the number of kick samples to be collected. This change is necessary to sample approximately the same area (1 square meter). Since the D-net is  $\approx 0.33$  m wide, we will sample a square area in front of the net of  $0.1108 \text{ m}^2$  ( $0.333\text{m} \times 0.333\text{m}$ ). In order to sample  $1 \text{ m}^2$ , we need to collect from 9 locations ( $0.1108 \text{ m}^2 \times 9 = 0.9972 \text{ m}^2$ ).

## **III. D-net – Multi-habitat Approach (Low Gradient Streams, Glide/Pool Habitat=Non-Comparable)**

The RBP procedures described above are only applicable to flowing, wadeable streams. The Multi-habitat Approach is based on protocols developed by the Mid-Atlantic Coastal Streams (MACS) Workgroup, which are employed in low gradient, slow moving streams. *This method is to be used only in wetland type habitat where flow is insufficient to move suspended materials into a net.*

Note: This type of sampling is considered non-comparable at this time as the

majority of other samples taken by the Watershed Assessment Section and analyzed using the WVSCI (West Virginia Stream Condition Index). Therefore, it should only be used for special surveys/projects or if specifically specified in the sampling plan/instructions.

1. Determine the types of productive habitat to be sampled and the percentage of each habitat within the sample station. Productive habitats are snags, vegetated banks, and submerged macrophytes. A total of 20 jab-sweeps (see next step) are collected based on the proportion of productive habitats available in the 100-meter assessment area. For example, if 50% of the habitat is snag material and 50% is submerged macrophytes, then 10 jab-sweeps (50%) are taken in snags and 10 jab-sweeps (50%) are taken in submerged macrophytes. If a particular type of habitat is rare (<5%), it is not sampled.
2. Collect macroinvertebrates by jab-sweeping the net into productive and stable habitat. A "jab-sweep" is an aggressive thrusting and sweeping of the net into productive habitat for a distance of one half meter. **Make only one jab-sweep; resist the urge to re-sweep!** A total of 20 jab-sweeps will be combined to complete the sample. The precise jab-sweep technique will vary with the type of habitat being sampled.
  - A. *Snags* –Disturb the snag area first by kicking it to dislodge the organisms. Then quickly jab-sweep the net into small sticks and branches or scrape the net along the lower surface of logs. Medium sized snag material is best –sticks and branches. Large logs should be avoided because they are generally difficult to sample adequately.
  - B. *Submerged Macrophytes* - In deep water, drag the net through the vegetation from the bottom to the water surface (maximum of 0.5 m each jab). In shallow water, bump the net along the stream bottom within the macrophyte bed, avoiding sediments where possible.
  - C. *Vegetated and Undercut Banks* - Use the snag collection method for collecting from roots and emergent plants that are on the lower banks of streams. Submerged areas of undercut banks are included here. Sample unvegetated banks by bumping the net along the substrate.
3. After five jab-sweeps have been collected, empty the net into a 5-gallon bucket containing stream water. (The net may be emptied more frequently, depending on the amount of material.) Repeat until 20 jab-sweeps have been collected.

The remaining procedure is the same as for the Rectangular Dipnet. Follow steps 6 through 12 under Sample Collection Methods – I. Rectangular Dipnet (Riffle/Run Habitats = Comparable) to complete field processing and preservation.

#### **IV. Hand Picking (Small narrow streams with minimal/interstitial flow = Non-Comparable)**

This sampling method should only be used for special surveys/projects or if specifically specified in the sampling plan/instructions as it is considered non-comparable to other samples. This method should be used in very shallow low-flow situations where there is not enough water to flow over the lip of the Rectangular Dipnet or D-net. Do not collect a sample if there is no interstitial flow in the areas between pools.

1. Sample in areas that would be considered riffles in higher flows. Do not sample in pool habitat. Pick up rocks (small gravel to small boulder) from about 0.25 m<sup>2</sup> (same area as that would be sampled by the Rectangular Dipnet) of substrate. Rub and rinse the rocks into a 5 gallon bucket partially filled with water. Repeat this procedure at four different areas - looking for the best habitats (highest interstitial water flow and most cobble sized rocks).
2. Use the rocks sampled to complete the benthic substrate section of the Habitat Assessment Form.
3. Pour the entire contents of the bucket through a U.S. Standard 30 sieve. Using a squirt bottle, rinse any remaining organisms from the bucket onto the sieve. Using forceps, remove any remaining organisms and transfer to jar. Place sample jar in cooler or other air-tight container designated for benthic macroinvertebrates.

The remaining procedure is the same as for the Rectangular Dipnet. Follow steps 7 through 12 under Sample Collection Methods – I. Rectangular Dipnet (Riffle/Run Habitats = Comparable) to complete field processing and preservation.

### **SAMPLE PRESERVATION METHODS**

1. Fill jar nearly full with 95% ethanol so that the concentration of ethanol is at least 70%. If there is a small amount of water in the sample, it may not be necessary

to fill the jar entirely to reach a 70% concentration. It is **very important** that sufficient ethanol be used to reach 70% concentration. In addition, enough alcohol should be added to at least immerse all of the material in the jar.

2. Make sure that a *label for the inside of the jar* in pencil (ink will run), include stream name, ANcode, and date. Place the label inside of the sample jar. Place the jar in a cooler or other container designated for the storage of benthic macroinvertebrates.
3. Try to keep from shaking the jars as much as possible. **Never invert the jars.**

## **LABORATORY METHODS**

Upon return to the office, all samples are to be logged into the Macroinvertebrate Sample Logbook. Each entry is to include: Date of Collection, date received by office, stream name, Random number (if applicable), AN-Code, and collector's initials. If a sample is in multiple jars, each jar is entered individually and designated as "1 of 2" or "2 of 2", as appropriate.

All macroinvertebrate samples are returned to the lab in-house sorting and identification or shipped to a contractor for sorting and identification. Sorting is done utilizing a modification of the RBP II 200-count sub-sampling method. Benthos are identified to the family level (in-house) or genus level (contractor) and the functional feeding group and tolerance values are determined. See SOP H-Processing Macroinvertebrate Samples and SOP I- Identification of Macroinvertebrates, Taxonomic References, and Data Analysis for more details about these processes.

## **QUALITY ASSURANCE/QUALITY CONTROL**

Replicate sampling is performed at 2.5% of the sites. Sites to have replicate macroinvertebrate samples are randomly selected via computer program. Replicates are obtained at the same time as the original sample. Different individuals collect samples so that the variances between individual techniques can be documented. Care is taken to assure that the replicate is not taken from an area that may have been depleted by previous sampling.

## **SOP H. PROCESSING MACROINVERTEBRATE SAMPLES**

### ***MATERIALS AND SUPPLIES***

1. Sample Jar - Contains the unprocessed sample.
2. Sample Bottle - for storage of processed sample.
3. Enamel Pans - contains sample during the sorting process.
4. Denatured Alcohol - preservative used in unprocessed and processed samples.
5. Sieves - #30 sieves are used to separate alcohol and fine debris from the sample prior to picking.
6. Sieve box - a home-made wooden frame with #30 mesh screening on the bottom is used to evenly distribute the sieved sample for randomly selecting the sub sample. The internal dimensions of the box are 10 inches by 10 inches. The screening is marked into 100 1 inch X 1 inch grids. This defines the sub sample to be picked.
7. "Cookie Cutter" - a homemade cookie cutter, 1 inch by 1 inch is used in conjunction with the sieve box to isolate each of the sub samples.
8. Labels - Self-adhesive labels are used to identify the contents of the sample bottle (i.e., the picked sample).
9. Scotch Tape - Used on label as additional adhesive.
10. Pencil - used to label sample bottle.
11. Crucible - or other small container, is used for short term, intermediate storage of the sample during the picking process.
12. Forceps - Fine tipped forceps are used to remove the organisms from the debris.
13. Illuminated Magnifier - an optical aid to illuminate and magnify the sample during the picking process. Alternatively, magnifying visors and a desk lamp can be used.
14. Squirt bottle - filled with alcohol, used to rinse organisms into sample bottle.



15. Plexiglas - used to cover sample overnight to prevent evaporation.

## ***SAFETY PRECAUTIONS***

Protective eyewear should be worn during sample processing to prevent contact with the residual alcohol in the specimens and debris.

## ***INTRODUCTION***

All macroinvertebrate samples are returned to the lab in-house sorting and identification or shipped to a contractor for sorting and identification. Sorting is done utilizing a modification of the RBP II 200-count sub-sampling method. Benthos are identified to the family level (in-house) or genus level (contractor) and the functional feeding group and tolerance values are determined. See SOP I- Identification of Macroinvertebrates, Taxonomic References, and Data Analysis for more details about benthic macroinvertebrate identification.

Sorting macroinvertebrates from benthic survey samples (a procedure often referred to as "bug picking") is an extremely important step in the biological research performed by the Department of Environmental Protection. The quality of the work performed by the "picker" influences the quality of subsequent processes, such as identification and data analysis. A competent "picker" must be able to recognize the morphological diversity of aquatic organisms, as well as the various methods these organisms may use to hide themselves from predators. The outcome of the final study may be affected, even if only a few organisms are overlooked during the picking process.

The biologists at the Department of Environmental Protection acknowledge the fact that the sorting process can be tedious at times. The picker is advised to discuss alternate sorting techniques that may be applied to difficult samples with senior biologists.

## ***DEFINITIONS***

**MACROINVERTEBRATES** - Animals that are large enough to be seen with the naked eye and do not have a backbone.

**BENTHIC ORGANISMS (or BENTHOS)** - Living organisms that reside on the bottom of streams, rivers, or lakes. Benthos may be vertebrates, invertebrates, or plants.

KICK - One method for collecting benthos. A hand-held net is held in the stream. The stream bed upstream of the net is disturbed using a kicking motion to dislodge the organisms, which then float into the net.

## **PROCEDURES**

1. Select the sample to be picked. A supervising biologist may provide the picker with a particular sample, or set of samples, that is to be picked.
2. Select a small bottle that will hold the organisms after sorting is completed. Usually 10 mL bottle is adequate for a 200-organisms sub-sample. A larger bottle may be needed if the sample contains large organisms.
3. Label the bottle:
  - a. Use self-adhesive labels
  - b. Using a pencil (ink will run if alcohol is spilled on the label), copy all information on the sample jar label onto the self-adhesive label. The label must include the following information:
    - ✓ Stream Name
    - ✓ Station Number (Random Number and/or AN-code)
    - ✓ Sample Date
    - ✓ County
    - ✓ Collection Method
    - ✓ Initials of Sample Collector
    - ✓ Initials of Sample Processor
    - ✓ # of grids picked
    - ✓ # of organisms in final sampleIf any of this information is missing from the original sample jar label, notify the supervising biologist so that the error can be corrected.
  - c. Stick the new label on the bottle and secure with scotch tape.
4. Prepare the sample for picking. This step is performed in a sink and should be done under a fume hood or in a well ventilated area.
  - a. Under a fume hood, open sample jar and pour contents into the # 30 mesh sieve.
  - b. Rinse sample jar into sieve and examine jar to make sure all detritus has been removed.
  - c. Rinse the contents of the sieve in tap water to remove remaining alcohol and

to rinse out fine sand and sediment.

- d. Carefully rinse any large detritus (i.e. leaves) or stones, making sure that all organisms on these items are returned to the sieve. Discard the leaves and rocks after rinsing.
- e. Place the contents of the sieve in the gridded screen box. Place the box in a few inches of water and gently swirl it until the contents are evenly distributed. If the sample was divided into more than one jar, the jars are to be combined at this point. When the sample is evenly distributed through out the gridded screen box, remove it from the water.
- f. Using a random number generator, select the first grid to be picked. Using the "cookie cutter", isolate the organisms within the chosen grid and scoop the contents of the grid into a white enamel pan. Be careful not to destroy any organisms during this step. Organisms with their head inside the grid are to be included within the grid. If you can't tell which end is the head, then the organism belongs in the grid that contains the largest portion of the body.

## 5. Picking

- a. Fill a crucible with 75% alcohol. A small piece of tape, rolled into a ring so the adhesive is exposed, may be attached to the bottom of the crucible to prevent tipping. If preferred, another small wide-mouth container may be substituted for the crucible.
- b. Using fine-tipped forceps and illuminated magnifier or magni-visor, remove all invertebrates from the sub-sample and transfer to the alcohol filled crucible. Keep track of the number of organisms that have been picked.
- c. If leaves are present, be sure to examine both surfaces. Watch for unusual clumps of twigs, leaves, or sand, which may be protective cases for some organisms. If cases are found, both the case and the organism should be picked. If the organism is in the case, the case and organism should be kept together. If an empty case is found, it should also be removed.
- d. If there is any doubt to the identity of an object (is it a seed or a bug?), it should be picked, but not counted. A senior biologist should be notified if a large number of questionable objects are present.
- e. When all the organisms appear to have been removed from the pan, agitate

the contents of the pan and look again. Often the agitation will reorient an organism that was previously overlooked.

- f. Have a senior biologist inspect the pan after picking has been completed. The biologist will point out any organisms that have been overlooked or misidentified as detritus. As the picker becomes more proficient at his/her task, this step will be reduced in frequency.
- g. Discard the contents of the enamel pan by pouring the contents through a "waste sieve" in the sink. The contents of the waste sieve may be emptied into the trash as necessary.
- h. If 200 or more organisms have been obtained from the initial grid chosen, sub-sampling is complete. If fewer than 180 organisms have been collected, another grid is randomly chosen and steps 4.f through 5.e are repeated until at least 179 organisms are obtained or until the entire sample has been picked.
- i. Pour the contents of the crucible into the labeled bottle. Use a squirt bottle containing alcohol to rinse the organisms from the crucible. Make sure that all organisms in the bottle are fully submerged in the alcohol and that none are clinging to the sides of the bottle. Use the squirt bottle to rinse the sides of the bottle, if necessary.
- j. If required, return the remainder of the unpicked sample to the original sample jar and preserve with alcohol. These samples will be processed to determine picking efficiency.

## 6. Record Keeping

After a sample has been picked, record the date and your initials in the sample log book.

## **QUALITY ASSURANCE/QUALITY CONTROL**

Sorting efficiency is evaluated for 2.5% of the samples. These samples are randomly selected. Pickers are instructed to retain both the picked portion and unpicked portion in separate containers. The unpicked portion is checked by senior biologist to determine that all organisms have been picked (recovery errors). The picked portion is sorted by a senior biologist to evaluate the picker efficiency.



# **SOP I. IDENTIFICATION OF MACROINVERTEBRATES, TAXONOMIC REFERENCES, AND DATA ANALYSIS**

## ***MATERIALS AND SUPPLIES***

1. Dissecting Microscope - for examination of gross features.
2. Compound Microscope - for examining minute features.
3. Fine-tipped forceps - for manipulating specimens.
4. Fine-tipped probes - for manipulating specimens.
5. Petri dishes - hold specimens during identification.
6. Alcohol - 75% ethanol or isopropanol is used to preserve the samples and to prevent desiccation during identification.
7. Wash Bottle - used for alcohol storage.
8. Microscope Slides and cover slips - for examination of tiny specimens and/or body parts under a compound microscope.
9. Benthic Macroinvertebrate Lab Sheet - standard for recording results of identification and enumeration (See Figure I-1).
10. Taxonomic Keys (see below)

## ***INTRODUCTION***

All macroinvertebrate samples are returned to the lab in-house sorting and identification or shipped to a contractor for sorting and identification. Sorting is done utilizing a modification of the RBP II 200-count sub-sampling method. Benthos are identified to the family level (in-house) or genus level (contractor) and the functional feeding group and tolerance values are determined.

## ***MACROINVERTEBRATE IDENTIFICATION PROCEDURES***

1. Check out the sample to be identified by completing the "WAP Bug ID" Log. Write down the date the sample was collected, streams and county, AN-Code, sign-out date, and your initials.
2. Complete the top portion of a "Benthic Macroinvertebrate Lab Sheet" (See Figure I-1).
3. Using the taxonomic keys listed above, identify the contents of the sample to the family level, or the lowest possible taxon. Use the reference collection as

additional confirmation, if necessary. **IF YOU HAVE ANY UNCERTAINTY ABOUT THE IDENTIFICATION OF A SPECIMEN, CONSULT A FELLOW BIOLOGIST FOR CONFIRMATION.** If an organism is too small or damaged and cannot be identified to the family level, identify it to the lowest positively-identified taxon and document why the identification was not complete.

4. Record results of identification and enumeration on a "Benthic Macroinvertebrate Lab Sheet" (See Figure I-1).
5. When the entire sample has been identified, submit the Lab Sheet to Janice Smithson or John Wirts.
6. Return the specimens to the original sample bottle and mark the label with an "X" to indicate the sample has been identified.
7. Place the sample in the box designated for identified samples and indicate that the sample has been returned by filling in the "Date In" section of the "WAP Bug ID" log.





## **TAXONOMIC REFERENCES**

The taxonomic references most frequently used by the WAS biologists for identification of macroinvertebrates are as follows:

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## **MACROINVERTEBRATE DATA ANALYSIS**

### **1. WVSCI**

The following metrics are applied to the benthic data:

1. Taxa Richness
2. Ephemeroptera, Plecoptera, Trichoptera (EPT) Taxa
3. Percent EPT
4. Percent Contribution of Dominant 2 taxa
5. Percent Chironomidae
6. Modified (family level) HBI (Hilsenhoff Biotic Index)

The individual metric scores are then standardized on a 100 point scale based on best standard values for a set of reference sites or conditions. The scores are then averaged to give the WVSCI (West Virginia Stream Condition Index).

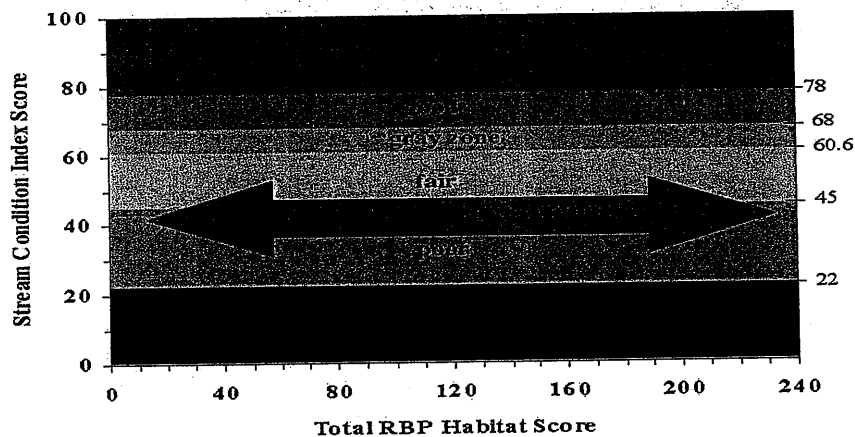
Macroinvertebrate data is evaluated through the preparation of a stream assessment chart (Figure I-2). This chart considers the biological and habitat conditions of each stream and compares them to those of the reference sites. Reference sites are those stations having optimal habitat (as defined by the RBP/EMAP matrix scores) and no obvious impairments in water quality. The number of reference sites selected depends on such variables as stream order and ecoregions. The framework for these assessments is the West Virginia Stream Characterization Index (WVSCI). Tetra Tech, Inc. (1), developed this index specifically for use in West Virginia. Stream scores are plotted within this chart and the results are used for overall watershed assessments, 305(b) reporting and 303(d) listing. Streams falling in the green area are considered fully supporting (for 305(b) reporting) or non-impaired (for WAP reporting). The condition of streams in the gray

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(1) Tetra Tech, Inc. A Stream Condition Index for West Virginia Wadeable Streams. July 21, 2000. Red Run Boulevard, Suite 110, Owings Mills, MD, 21117.

area may be fully supporting or threatened (305(b)). Water quality data must be evaluated to determine if a stream in the gray area is threatened or fully supporting. Often best professional judgment cannot be avoided. The yellow area contains streams that are partially supporting (305(b)) or impaired (WAP). Streams in the orange and red sections are non-supporting (305(b)) or impaired. All streams falling in the yellow, orange and red sections are subject to inclusion on the 303(d) list.

**Figure I-2: Scoring Categories for WAS Assessed Streams**



## 2. Dirty Null Stressor Identification Model

The benthic data is also imported into an analysis model that compares each sample's community structure to that of a set of "reference" data with well known and established stressor types (Fecal, Metals, Sediment, Sulfate, and Reference Condition), also known as "Dirty Nulls". The data that results from the Dirty Null Stressor Identification Model is a set of similarity indexes and probability percentages that help identify potential stressor or stressors to the stream community.

## **INTEGRATION OF BIOLOGICAL, HABITAT, AND WATER QUALITY DATA**

The WVSCI scores and Dirty Null Stressor data are compared to what is known about the habitat and water quality at each site in order to understand why the stream scored the way it did. Those sites having significant impairment are subjected to more intensive surveys in the future.

## **QUALITY ASSURANCE/QUALITY CONTROL**

The integrity of the taxonomist plays an important part in the accuracy of identification. All taxonomists are encouraged to consult their peers when a difficult or unusual specimen is encountered.

A reference collection is being compiled to serve as a resource to all taxonomists. All organisms in the 200-specimen sub-sample are retained in a voucher collection.

Identifications are verified for 2.5% of the samples by the senior biologist. Samples for reidentification are selected randomly and will encompass checks on all taxonomists.

## **SOP J. PERIPHYTON**

### **MATERIALS AND SUPPLIES**

1. Support Ring – A piece of PVC pipe (1 cm long & 4 cm inside diameter) to delimit the sample area on rocks (12.56 cm<sup>2</sup> of area inside ring)
2. Scraping tool (microspatula or knife)
3. Small brush (toothbrush) that is replaced weekly
4. Sample container (4 oz “specimen jar”)
5. 10 % formalin (for sample fixing/preservation)
6. Cooler w/ ice (for sample storage/preservation)
7. Electrical tape (for sealing lids of sample jars)
8. Labels (labels are to be placed inside sample container & on outside)
9. Clear tape (to affix label to container)
10. Squirt Bottle

### **SAFETY PRECAUTIONS**

Rubber gloves and protective eyewear should be worn during sample collection to avoid bacterial contamination. They should also be worn during sample preservation or at any time while handling formalin.

### **INTRODUCTION**

Periphyton are attached algae, i.e., algae that grow on the exposed surfaces of rocks and other submerged objects. Phyto-benthic (bottom-dwelling) algae are usually the dominant component of a periphyton community. Phyto-benthic algae, the primary producers in the stream ecosystem, are sensitive indicators of change in lotic waters. Because it is attached to the substrate, the periphyton community integrates physical and chemical disturbances to a stream. Another advantage of using periphyton in water quality assessments is that the periphyton community contains a naturally high number of species, making data useful for statistical and numerical applications to assess water quality. Response time of the periphyton is rapid, as is recovery time, with recolonization after a disturbance often more rapid than for other organisms. Diatoms, in particular, are useful indicators of biological integrity because they are ubiquitous; at least a few can be found under almost any conditions. Most diatoms can be identified to species by experienced biologists and tolerances or sensitivities to specific changes in environmental conditions are known for many species. By using algal data in association with macroinvertebrate data, the biological integrity of stream ecosystems can be better ascertained.

## **SAMPLING PROCEDURES**

1. Collect periphyton at all reference, random sites, TMDL Bio Sites, and those non-random sites with nutrient enrichment indicators or as directed on the stream list. Periphyton may also be collected at big streams (large rivers where WVSCI is not applicable – Elk River near mouth), and at streams that are too deep for benthos collection (i.e., water over the net) but not too deep to reach in and grab cobble to sample periphyton as a biological indicator for the site.
2. Ideally, samples should only be collected during stable flow conditions. After extremes of flooding or drought, a two-week period is required for adequate recolonization. Because WAP sampling is conducted within a short index period (random sites), periphyton will be collected when streams are not turbid (i.e., the substrate is visible).
3. Label sample container with Stream Name, AN-Code, date, collector, and “w/ formalin”.
4. To be consistent, samples will only be collected from rocks (epilithic habitat) from riffle/run areas of the streams. Collect five cobble-sized rocks that are exposed to light from throughout the reach. If there are no rocks available from the reach, collect periphyton from removable wood (same technique as for rocks), documenting on the field sheet exactly what was sampled. These riffle/run areas should coincide with the areas where benthic macroinvertebrates are collected (if benthos collected). Using the PVC ring to delimit the sample area (12.56 cm<sup>2</sup>), scrape **all algae** from **upper surface** of rocks into the sample jar. Use the toothbrush to loosen any remaining periphyton. Remove sampler and rinse loosened algae into the sample jar using clear stream water collected from that site in the squirt bottle. Periphyton from the five rocks (representing 62.8 cm<sup>2</sup> of sampled area) is composited into one sample jar. Snap the labeled lid onto the container.
5. Rinse the sampler and brush and/or scraping device thoroughly with stream water at the site before and after each sampling event to avoid contamination of subsequent collections.
6. A guideline for preservation is as follows: Assuming the sample jar is about 3/4 (120 ml) full, preserve with an adequate amount (a “plop”) of 10% formalin from the squeeze bottle) for sparse to normal periphyton amounts. Add more for samples with heavy amounts of green algae. The specimens cannot be over preserved. The specimen cups are graduated (ml) so

adding the proper amount of formalin can be measured. Take extra care when preserving, as formalin is a known carcinogen. Note: Samples do not need to be preserved immediately. It is probably easier to preserve all periphyton samples collected in a given day at one time – upon returning to office or hotel parking lot. Whether samples are fixed immediately or not, they should be placed in a cooler with ice. Sample jars should be taped by sealing the rim of the lid to minimize the chance of spillage.

7. Record the number of rocks “scraped” and comments about where the rocks were collected, such as primarily exposure to sunlight (ex. 3 rocks with full exposure, 2 with partial shade). The yes/no questions and comments box in the Habitat Assessment Form (see SOP B- Instructions for Completing the Habitat Evaluation Form) will be used to aid in interpreting data from scoured or drought affected reaches.

## **LABORATORY METHODS**

Periphyton identification and biomass determinations are performed by a private contractor. The contractor is required to have a degreed biologist on staff that performs the actual identifications. The contractor must adhere to the following protocols.

- A. **“Soft” Algae (Non-Diatoms)** – Relative and abundance are to be determined as follows:

Homogenize sample in a blender and pipette a subsample into a Palmer counting cell. Dilute the sample if cells overlap too much for accurate counting. Identify and count 300 non-diatom algal units to the lowest taxonomic level at 400X magnification. Colonial species are to be counted as individual cells, when appropriate. Filamentous species should not be counted as individual cells, but as cell units of 10 micrometers in length. The number of “live” diatoms observed should also be recorded (identification will be done under a separate procedure). Record the numbers and species of “soft” algae on the bench sheet.

- B. **Diatoms** – Diatoms are to be analyzed after the “soft” algal identifications are complete, as the clearing process will destroy soft tissue. Procedures are as follows:

Clear diatom frustules of organic material using either nitric acid or hydrogen peroxide/potassium dichromate oxidation. Prepare slides and identify diatoms to species or lowest taxonomic level possible. Record all taxa encountered on the bench sheet to create a species list prior to enumeration. Continue identification until no new taxa are found after a 2-3



minute scan. To obtain quantitative data, count a minimum of 600 valves and record the taxa and number encountered on the bench sheet.

## **DATA ASSESSMENT**

An assessment of biological integrity can be made based on the periphyton data. The goal is to categorize water quality as excellent, good, fair, or poor and to determine the degree and cause of aquatic life use impairments in fair or poor streams.

Biological indices represent mathematical models of community changes. Changes in water quality will affect resident biota, and indices that reflect these changes in a particular community are useful biological indicators of water quality. The periphyton community, especially diatoms, is a useful biological indicator because:

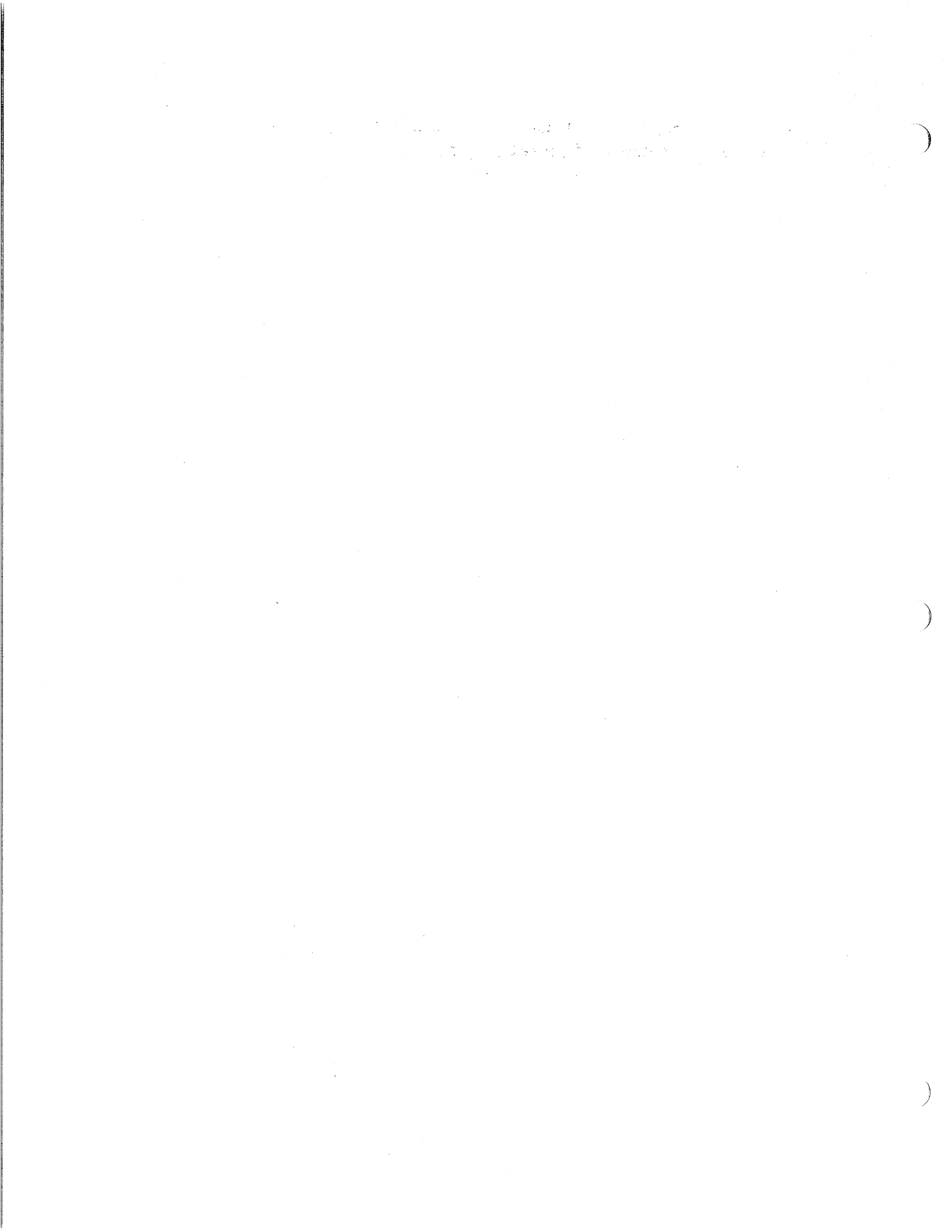
- They are attached to the substrate and, therefore, subjected to any immediate or prolonged disturbances;
- Diatoms are ubiquitous, with at least a few individuals found under almost any aquatic conditions;
- Total number of taxa at any given site is usually high enough for use in calculating various metrics;
- Diatoms, especially the most abundant species, are identifiable to species by trained professionals;
- Tolerance of or sensitivity to changes (autecological requirements) is known or suspected for many species or assemblages of diatoms; and
- Periphyton communities, especially diatoms, have a rapid response and recovery time because of their relatively short lifecycle (as compared to fish or macroinvertebrates) and their ability to quickly recolonize formerly disturbed (impacted) sites.

Several metrics have been used successfully to assess water quality conditions using periphyton. Some have the diagnostic ability to indicate the type of impact (nutrient enrichment, toxicity, acidity, salinity, sewage (organic) pollution, and siltation).

## **QUALITY ASSURANCE AND QUALITY CONTROL**

Sample labels are to be accurate and complete and contain all the information discussed in Section 3-C of this document. Sampling equipment will be checked for residual algal material, rubbed clean and thoroughly rinsed with stream water before and after each sampling event. Duplicate samples will be collected from 10% of the sites sampled. Periphyton will be collected along with other activities at

the designated duplicate WAP sites. Duplicate samples will be analyzed to ensure precision and repeatability of the sampling technique.



## SOP K. MEASURING STREAM FLOW (INCLUDING OPERATION AND CALIBRATION)

### **MATERIALS AND SUPPLIES**

1. Wading Rod – for measuring stream depth and setting depth of flow measuring device.
2. Marsh-McBurney Flo-Mate – for measuring water velocity.
3. Tape Measure in feet and tenths – for determining the distance between velocity readings.
4. Flow Record Sheets (included in Habitat Assessment Form) – for recording data collected along the transect and for final computation of flow.
5. Pencils.

### **INTRODUCTION**

Most discharge measurements of stream flow are made by the current-meter method because it is adaptable to a wide range of velocities and is practically unlimited as to the total discharge which can be measured. Essentially, the method consists of: 1) measuring the velocity of flow in and the area of each of several parts of a cross-sectional transect; 2) computing the discharge in each part as the product of the velocity and area; and 3) summing the partial discharges to obtain the total.

The usual method of making a discharge measurement is explained in Figure G-1, which shows the cross-section of a channel. The depth of water is measured by rod at location 1, 2, 3, 4, and so forth. The velocity of the water is measured by current meter at each of these locations at such position(s) in the vertical that the mean velocity in the vertical is obtained.

The discharge past a partial section is computed by:

$$q_4 = V_4 \frac{[(L_4 - L_3) + (L_5 - L_4)]}{2} \quad (d_4) = (V_4) \frac{[L_5 - L_4]}{2} \quad (d_4)$$

Where  $q_4$  = discharge in cubic feet per second through partial section 4 (Fig. G-1).

$V_4$  = mean velocity in feet per second in the vertical at location 4.

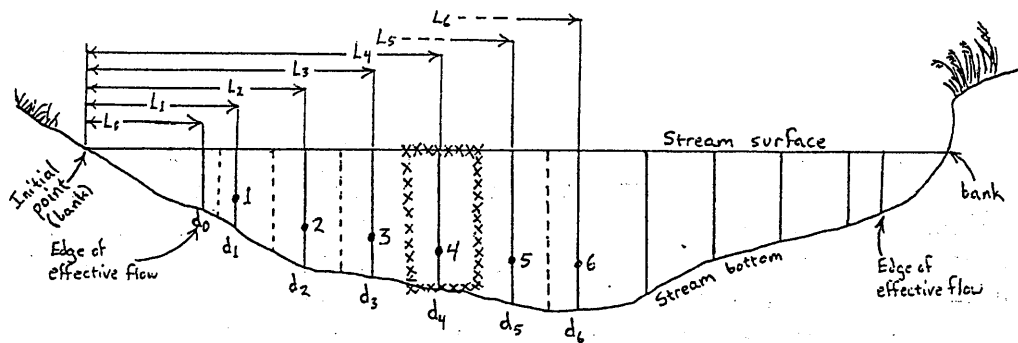
$L_3, L_4, L_5$  = distances in feet (not inches) from the initial point to locations 3, 4, and 5, respectively (Fig. G-1).

$d_4$  = depth of water in feet (not inches) at location 4.

The area which is defined by this formula is that shown by the X line around location 4 in Figure G-1.

The summation of the discharges for all the partial sections is the total discharge of the streams. This method of computation is fittingly called the SUM-OF-PARTIAL-DISCHARGES METHOD, an example of which is shown in Figure G-3. The calculation process is presented step-by-step in another section.

**Figure G-1. Cross section of stream channel**



1, 2, 3, 4, 5, 6, - Observation Points.

$L_1, L_2, L_3, L_4, L_5, L_6$  - Distance in feet from the initial point to the observation point for six consecutive partial sections.

$d_1, d_2, d_3, d_4, d_5, d_6$  - Depth of water in feet at the observation point in the partial section.

Dotted lines indicate the boundaries of a partial section.

## I. Operation and Care of Flo-Mate







The use of the function keys are described in Figure G-2.

Figure G-2. Key Function descriptions for the Cross section of stream channel









# FLO-MATE™

## Model 2000 Key Summary

### One Key Function

-  - Turns Unit ON. Clears the display and restarts the meter.
-  - Turns Unit OFF.
-  - Increments Fixed Period Averaging, Time Constant and Memory Location.
-  - Decrements Fixed Period Averaging, Time Constant and Memory Location.
-  - Switches Between Recall and Primary Operating Modes.
-  - Stores Values In Memory.

### Two Key Function (Press at same time)

-   - Change Units, Turns Beeper ON/OFF
-   - Toggles Between Fixed Point Averaging and Time Constant.
-   - Clears Memory. Meter must be in the primary operating mode.
-   - Initiates zero adjust sequence. (Zero stability is  $\pm 0.05$  ft/sec. See instruction manual for procedure.)

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P/N # 102000601

### *Zeroing and Setup*

Every month, the meter will need to be zeroed and set up for taking measurements.

1. Clean the sensor with soap and water.

2. Place the sensor in a five-gallon bucket of water. Sensor should be 3 inches away from the sides and bottom of the bucket. Make sure the water is not moving and wait 10 to 15 minutes. **DO NOT TAKE ANY READINGS WHILE WAITING**
3. Press STO and RCL keys at the same time. The unit will display a "3".
4. Use the "down" arrow to decrement to zero. The number "32" will be displayed.
5. Unit will decrement itself to zero and turn off. Calibration is complete.
6. Set up the unit to read in Ft/S by pressing ON/C and OFF keys simultaneously. Press these two keys until FT/S is displayed. You can choose to have the beeper on or off (watch for the little speaker symbol in the lower right-hand corner).
7. Set up the unit to read in 20 second intervals. Press the Up and Down arrow keys at the same time until the meter displays FPA (fixed point average). Press the up arrow or down arrow to set the time for 15 seconds.

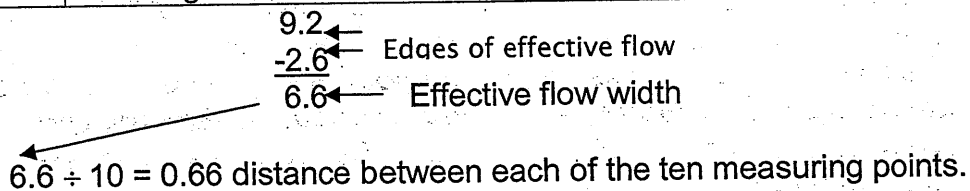
#### *Using the Flo-Mate*

1. Once calibration and initial set up are complete, **DON'T PRESS** any of the keys on the Flo-mate except the on OR off button. Pressing combinations of these keys will only mess up the set-up.
2. Turn the instrument on **AFTER** you have set up your flow transect and have the sensor in the water.
3. Allow the instrument to run through two cycles. During the first cycle, the readout will fluctuate while the sensor obtains data to average. The readout will be stable during the second cycle. At the end of the second cycle, record the readout. This readout is the average for the second 20 second cycle (the first 20 second cycle is to allow the turbulence and eddies around the flow rod and sensor to reach equilibrium).
4. Move the sensor to the second increment in the stream, wait for a **new** cycle to begin (you don't want the average to include movement while you were moving the wading staff. Wait for two complete cycles to pass and record the readout. Repeat until the transect is complete.

## II. FIELD PROCEDURES

Figure G-3. Example of Flow Measurement Form: Recording field data.

STREAM DISCHARGE MEASUREMENT (Calculated in cubic feet per second — cfs)					
Measurer	Doug Wood	Meter ID	2	Time	12 noon
Distance	Distance	Depth	Velocity	Discharge cfs	
Bank	1.3				
EEF	2.6	This is the Edge of Effective Flow			
1	2.93	0.64			
2	3.59	0.90			
3	4.25	0.93			
4	4.91	0.97			
5	5.57	1.01			
6	6.23	1.12			
7	6.89	1.06			
8	7.55	0.92			
9	8.21	0.88			
10	8.87	0.63			
11	9.2	0.60			
EEF	9.9	This is the Edge of Effective Flow			
Bank	10.5				
Notes:	Some turbulence from rocky substrate. Slightly skewed flow. Drizzling rain.				



**Note: ALL MEASUREMENTS TAKEN IN INCHES MUST BE CONVERTED TO FEET AND TENTHS/HUNDREDTHS IN ORDER TO GIVE A WIDTH MEASUREMENT THAT CAN BE USED IN CALCULATING FLOW IN UNITS OF CUBIC FEET PER SECOND (cfs).**

1. Select a stream reach having the following characteristics:
  - a. A straight stretch of water with the flow running parallel to the stream bank



- b. A stable, even streambed without large rocks, weeds and protruding obstructions that create turbulence.
- c. A level streambed configuration to reduce the vertical components of velocity.

2. All of these conditions are seldom satisfied. Select the best possible reach using these criteria, then select a transect.

An ideal transect:

- a. Is perpendicular to the direction of flow.
- b. Has uniform bed and stream banks
- c. Has a minimum velocity of 0.5 feet/second. (Avoid transects with eddies or areas of "dead" water.)
- d. Adequate depth for the meter to function.

**Note:** You may alter the channel at the selected site *before* you begin making any measurements, but **NEVER AFTER** measurement has begun.

3. Determine the width of the stream by placing a tape measure perpendicular to the stream flow.

4. Determine the width of "effective flow". The effective flow is the segment of the transect having measurable downstream velocity and exclusive of "dead" areas or reverse flows (eddies) beside the stream banks. Do not exclude similar anomalous flow areas that do not touch the banks from the width measurement. For simplicity, utilize this effective flow width in establishing endpoints for measurement of velocity and depth (See Fig. G-3).

5. Determine spacing of velocity measurements.

There is no set rule about how to space the measurements other than there is a required minimum number of measurements depending on the width of the effective flow. ***Velocity measurements should be spaced to document and define areas of turbulence, extreme changes in velocity, and sudden changes in depth. For example, if there is a large obstacle to the flow like an unmovable boulder directly in the flow transect, a measurement should be taken on both sides of the boulder and on top or behind it. The flow measurements can also be spaced farther apart in areas where the velocity and depth are more uniform.***

- a. In streams with effective flow width greater than 3 feet, take no fewer than ten nearly even-spaced measurements within the effective flow transect. For example, if the effective flow is 3.5 feet wide, measurements could be taken every 4.2 inches (this is 10% of the effective flow width, 42 inches).

- b. If the effective flow width is less than 3 feet, take as many measurements as possible using an increment no smaller than 0.3 ft using best professional judgment.
- c. If the effective flow is >10 feet, a minimum of 20 measurements should be obtained.

6. Record the following information on the Discharge Measurement section of in the Habitat Assessment Form:

- a. Record the flow measurer.
- b. The time of the flow measurement.
- c. The assigned number of the flow meter used.
- d. Any conditions that might affect the flow measurement (wind, rain, skewed bottom configuration, ice and leaf packs in the water).

7. Begin Flow measurement:

- a. For simplicity, establish the bank as the initial point. Then record the distance ( $L_0$ ) from the initial point to the edge of effective flow. Record the depth at the edge of effective flow ( $d_0$ ).
- b. Record the distance ( $L_1$ ) from the edge of effective flow to the first velocity measuring point. (This point is called a "vertical"). This first vertical may be established at a distance close to the edge of effective flow (about 0.3-0.5 feet). Record water depth ( $d_1$ ) at this vertical.
- c. If the depth is less than 2 feet, use the one-point 0.6-depth method for measuring velocity: Adjust the rod so that the sensor is at the depth that is six-tenths below the water surface (indicated by "1" in Figure G-1). If the depth is greater than 2 feet, two readings are taken; 0.2 and 0.8 from the surface.
- d. Place the sensor at the proper depth and allow it to adjust to the current before starting the observation. The following precautions should be taken:
  1. Hold the rod straight up with the sensor pointing upstream.
  2. Stand downstream from the meter and at a distance so that you do not create turbulence that will impact the reading. In very small streams, attempt to straddle the wetted area to avoid altering the flow.
- e. Allow the Flo-Mate to go through two complete cycles (40 seconds). Record the second readout on the "velocity" column on the form.

8. Repeat Step 7 at each vertical. Be sure to record the distance to and depth of the far point of effective flow, and the distance to the bank.

### III. Calculating Flow

#### *Computer Calculation of Flow*

1. Open Excel, then open the file in G:/WAP/Important Wap Files/Flow Template.xls.
2. At the bottom of the spreadsheet select the tab for the number of actual flow readings obtained. If there is no tab for the number of measurements you have taken, you will need to insert a new worksheet, copy contents of another worksheet into the new worksheet, and add or delete rows as needed.
3. Type in the left and right edge-of-effective-flow values in the "no flow" rows of the "Distance" column.
4. Enter the distance, depth, and velocity in the appropriate columns.
5. The flow or discharge in CFS is automatically calculated in the lower right-hand corner of the spreadsheet.

#### *Manual Calculation*

**Step 1.** The first step is to determine the distance between each partial section. A partial section is illustrated in Figure G-1. To do this you must first determine the width of the verticals. A column has been added to the flow measurement table to illustrate how verticals widths are determined. The table in Figure G-4 provides details for the examples below.

To determine the first vertical width, subtract distance 1a (See Fig. G-4) from distance 1b and divide by 2:

$$2.6 - 1.3 = 1.3$$

$$1.3 \div 2 = 0.65 \text{ the width between the first vertical (2b in table)}$$

Now, determine the width between the second two verticals:

$$\frac{1c - 1b}{2} \text{ OR } \frac{2.93 - 2.6}{2} = 0.165 \text{ The width between the second two verticals (2c)}$$

The sum of these two verticals will give you the width of the first partial section:

0.65 + 0.165 = 0.815 This result will go into the table at 3b.

Repeat this step until you have calculated all the partial section widths.

**Step 2:** Determine the individual flows for each partial section:

Within each row multiply width by depth by clicks and divide by seconds; for example in row c:

$$\frac{(0.495)(0.64)(4)}{62.3} = 0.02034$$

The flow for that partial section. Enter this number in 8c in the table.

Repeat Step 2 until calculations for each row have been made.

**Step 3.** Add all the numbers in column 8 get the total discharge in cfs.

**Figure G-4. Example of Flow Measurement Form: Calculating flow.**

Distance	Vertical Width	Width	Depth	Velocity	Discharge cfs
1a. 1.3		3a. Bank			
1b. 2.6	2b. 0.65	3b. 0.815	4b. 0.41	Outer edge of dead zone	
1c. 2.93	2c. 0.165	3c. 0.495	4c. 0.64		8a. 0.02034
1d. 3.59	2d. 0.33	3d. 0.66	4d. 0.90		8b. 0.18939
1e. 4.25	2e. 0.33	3e. 0.66	4e. 0.93		8c. 0.26854
1f. 4.91		3f. 0.66	4f. 0.97		8d. 0.34814
1g. 5.57		3g. 0.66	4g. 1.01		8e. 0.31555
1h. 6.23		3h. 0.66	4h. 1.12		8f. 0
1i. 6.89		3i. 0.66	4i. 1.06		8g. 0.03199
1j. 7.55		3j. 0.66	4j. 0.92		8h. 0.34180
1k. 8.21		3k. 0.66	4k. 0.88		8i. 0.22126
1l. 8.87	2l. 0.33	3l. 0.495	4l. 0.63		8j. 0.06575
1m. 9.2	2m. 0.165	3m. 0.515	4m. 0.60	Outer edge of eddy	
1n. 9.9	2n. 0.35	3n. Bank			
				Discharge =	1.80276 cfs

## **QUALITY ASSURANCE/QUALITY CONTROL**

Before use, each current-meter should be examined for wear and adjustments should be made as required. The rotor should spin freely and for the proper duration.

All individuals conducting field work will receive hands-on training prior to the field season. Individual training will occur simultaneously on the same stream so the results can be compared to the group average. Readings that deviate exceptionally from the norm will be examined for errors. In the field, flow-measurement teams will consist of two people. Individuals who are more experienced in determining flows will be teamed up with the less experienced to assure reinforcement of training and accurate results.

## **SOP L. SUBSTRATE CHARACTERIZATION (INCLUDING GRADIENT)**

### ***MATERIALS AND SUPPLIES***

1. 100 meter Measuring tape – used to delineate the length of the substrate characterization reach and to demarcate the data points along this reach.
2. Flagging tape – used to mark the eleven data point intervals in the substrate transect.
3. Survey Extension Pole – used to determine stream width and data points along each transect. Also used in conjunction with the clear plastic tubing to measure the rise in the stream between the two ends of the reach.
4. Thalweg Pole – used to measure the thalweg and to determine the substrate character at each transect data point. Also used in conjunction with the clear plastic tubing to measure the rise in the stream between the two ends of the reach.
5. Handheld Eye Level – used as an alternative method to measure the rise in the stream between the two ends of the reach.
6. Water Level – made of clear plastic tubing with valves on each end; used to measure the rise in the stream between the two ends of the reach.
7. Habitat Form – Forms for substrate characterization is a component of the WAS habitat sheet.

### ***PROCEDURES***

#### **I. Establishing Reach and Transects**

1. Determine the substrate characterization reach by multiplying the average stream width (as determined during the Rapid Bioassessment Protocol survey) by 40. The minimum and maximum widths are 100 and 500 meters, respectively. Record the reach length on the Habitat Assessment Form (See SOP-F Habitat).
2. Determine the transect intervals by dividing the total reach length by 10. Measurements are taken at each of these transects including the upstream and downstream endpoints for a total of 11 transects. Each transect is assigned a letter, with the first (downstream) transect identified as Transect A and the upstream terminus being Transect K.

#### **II. Substrate Measurement (AKA Pebble Count), Thalweg Profile, and Bankfull Height**

1. Begin at Transect A and work upstream. Mark the measurer and recorder of the data.

2. Using the survey extension pole, determine the wetted stream width. Divide the stream width by four to determine the measurement points. Measurements will be taken at the right descending bank (0% of the wetted-width), right-center (25% of the wetted-width), center (50% of the wetted-width), left-center (75% of the wetted-width), and at the left descending bank (100% of the wetted-width). **Note: If a split channel is encountered one of two things can occur:**

**A) If the split channel features a bar (*bar definition: a channel feature below the bankfull height that is dry during baseflow conditions*) then conduct the measurements at that transect as if there was only one channel and note the presence of the bar. Any measurements that fall on the bar should be treated just as if it was inundated with water, but noted as being taken on a bar.**

**B) If the split channel features an island (*island definition: a channel feature even with the surrounding flood plain or above the bankfull height that remains dry even at bankfull flow*) then conduct a separate transect in each channel for the length of the island. The situation should be documented and the second transect information is recorded continuing the transect letters down the alphabet starting with J.**

3. To take a substrate measurement, hold the thalweg pole vertically at the transect point and lower it straight down to the bottom. Pick up the particle at the tip of the pole (if it is not a boulder or bedrock). Using the markings on the thalweg pole, measure the particle at its median diameter. Each particle will have three dimensions: width, depth, and height. *Measure the "middle" dimension, i.e., the dimension that is neither the largest nor smallest.*

**NOTE:** In cases where there is a deposit of fine material (silt or sand) on top of another substrate type, you must use the THUNK test to determine which layer to count. The THUNK test consists of slowly lowering your thalweg pole straight down to the bottom as normal. If your pole hits the particle abruptly and makes a sort of "THUNK" sound, then the deposit of fine material is not considered and you count the underlying material. If your pole hits the bottom and can continue down to some degree with minimal resistance, then you record the fine material on top. Much of this determination relies upon experience and best professional judgment. Be sure to confer with your team partner and if in doubt, write notes.

Record the size class the particle falls into based on the following table:

Class	Size	Scor	Description
Bedrock (BR)	>4000 mm	6	Bigger than car
Boulder (BL)	>250-4000 mm	5	Basketball to car
Cobble (CB)	>64-250 mm	4	Tennisball to basketball
Coarse Gravel (GC)	>16-64 mm	3.5	Marble to Tennisball
Fine Gravel (GF)	>2-16 mm	2.5	Ladybug to marble
Sand (SA)	>0.06-2 mm	2	Gritty between fingers
Fines (FN)	<0.06 mm	1	Smooth, not gritty (silt & muck)
Hardpan Clay (HP)	>4000 mm	hp	Clay bottom
Woody Debris (WD)	Regardless of size	wd	Rootwads, snags, logs, sticks

Repeat this step for each of the five measurement points.

4. Determine the thalweg of the transect. The thalweg is the deepest part of the stream channel at the transect. Use the thalweg pole to determine water depth at the thalweg. **Read the depth on the *side* of the thalweg pole to avoid the wave produced by turbulence.**
5. The bankfull height is defined as the channel height that is filled by moderate-sized flood events that occur every one or two years. Look for a variety of bank characteristics to determine the bankfull height. Often there is an obvious slope break that differentiates the channel from a flat floodplain higher than the channel. A transition zone sometimes exists between exposed substrate and vegetation, which marks the bankfull height. Also, it may be determined by moss or vegetation growing on rocks along the banks. Sometimes the most obvious characteristic to look for is the presence of drift material (e.g., leaves, trash) along the bank or on overhanging branches. **Be sure to record a minimum of three bankfull height measurements throughout the sample reach. These measurements can occur anywhere along the reach, but should be spaced out along the reach.**
6. Repeat these steps for Transects B through K. The next transect can be located by moving upstream the transect interval as calculated above using the 100m measuring tape as a guide. Be sure the data from each transect is recorded in the appropriate space on the data sheet.

### III. Gradient Measurement

**IMPORTANT:** Gradient measurements must be taken along the full length of the reach. If the full reach is not measured with the tape measure, the gradient calculation will be incorrect and the time taken to record this data will be wasted.



There are two options for devices to measure slope: the Handheld Eye Level and the Water Level. Each device has its positives and negatives and each should be considered when selecting a device.

The Handheld Eye Level is much smaller than the Water Level in both weight and volume, so it may be more ideal in situations where a lengthy hike is necessary. It also is possible to measure longer distances with the Handheld Eye Level if the stream is straight enough and there are not major line-of-sight issues (*i.e.*, overhanging vegetation, houses, bends, etc.). Larger reaches (*e.g.*, >250 m) may benefit from the use of the Handheld Eye Level in both ease of use and reduction in the amount of time to obtain the Gradient Measured. On the negative side, the Handheld Eye Level is less accurate than the water level. It can also be problematic in raining conditions as the lenses can fog up. In addition, if two shorter people are working together, it may become necessary to use shorter distances between readings so that the person at the downstream end can point out with a stick or even read the level mark.

The Water Level is definitely more accurate. But the distance between readings is limited (usually to 20 m) by the length of tubing. In addition, high gradient streams may require one to shorten the length of the tube in order to capture the reading on the downstream end. The Water Level is ideal for situations with dense overhanging vegetation that prevent the use of the Handheld Eye Level. Because of its weight (the tubing and water inside the tubing) may be more useful when the sample site is immediately near the roadside and jeep.

### Measurement Methodology

- A. Handheld Eye Level Method - Slope is measured by "backsiting" or "backshooting" downstream between the two reach ends. The primary method is to use a handheld eye level. If a situation occurs where using the handheld eye level is not feasible (*e.g.*, the stream is too sinuous or there is too much overhanging vegetation) then the Water Level Method may be used instead to cover that distance.

**Note: Each individual should determine and remember their eye level height (the point on the survey pole that their eye is level with) before doing any slope measurements. Also, keep in mind that this point can change when wearing different wading boots or footwear.**

1. One individual stands at the water surface along the bank with the handheld eye level while the other holds the survey extension pole at the water surface downstream (as far as the individual can see accurately with the handheld eye level).

2. Looking through the eye level, the upstream individual determines where along the vertical surface of the survey extension pole their eye is level. The upstream individual will instruct the downstream individual to move a horizontal marker (e.g., finger, stick, pencil, thalweg pole) up or down to the same spot. The upstream individual's eye level height is then subtracted from the measured height to determine the rise of the stream for that distance. This value is recorded on the field form.
3. The upstream individual must now move to the position of the downstream individual (which can be marked by stacking rocks or with placement of an object or flagging) and the downstream individual moves down as far as the upstream individual can see accurately with the handheld eye level). Repeat the measurements as described until the downstream end has been reached.

B. Water Level Method - The secondary method is to use a water level in the form of clear plastic tubing with some sort of length measuring device on each end (e.g., the Thalweg and Survey Extension Poles). If a situation occurs where using the water level is not feasible (e.g., the stream goes under a road or culvert) then the handheld eye level may be used instead to cover that distance.

1. Fill the tubing by holding both ends level and pouring stream water collected in a cubitainer into the tubing until full. An alternative method to fill the tubing is to put stretch the tubing along the stream and submerge the upstream end under the water surface while siphoning the downstream end until enough of a draw is created to fill the tube.
2. Each partner secures an end by placing a rubber stopper or thumb into the end of the tubing, and then stretching the tubing to length along the contour of the stream starting at the upstream end of the reach.
3. Place the surveyor pole at water level at the downstream end. Stretch the tubing along the surveyor pole with the end of the tube at least to the 1 m mark (or higher if necessary). To help hold the tube against the pole, you may use your foot to help hold the tube at water level. Perform the same steps at the upstream end with the exception of using the thalweg pole.
4. When both ends are in position, the upstream individual must remove the stopper or thumb from the end of the tubing, followed by the downstream individual. The water level will oscillate until equilibrium is reached. Once the water level in the tube is stable, record the approximate location of the meniscus on each end, then subtract the

upstream from the downstream measurement. Record this as the change in elevation or rise of that stream segment.

5. The upstream individual must now move to the position of the downstream individual (which can be marked by stacking rocks or with placement of an object or flagging). The tubing is again stretched to length and the method is repeated. Repeat the measurements as described until the downstream end has been reached

**NOTE:** If you encounter a high waterfall in the reach, measure the rise from the edge (if safe) of the fall to the splash-zone below using the surveyor pole and record it in one of the extra blanks on the field form. Also describe the reading and include what it is (i.e. waterfall) and where in the reach the waterfall was located (transect location). Then continue measurements past the waterfall as normal.

The final gradient measurement (% Gradient) is calculated after data entry via a query calculation:

$$\% \text{ Gradient} = \frac{\text{Sum of Rises} \times .100}{\text{Reach Length}}$$

#### **IV. Final Result**

All of this data (Pebble Count, Thalweg Profile, Bankfull Height, and Gradient) are entered into the WAS database and numerous values and statistics are calculated via a series of queries. These values define the characteristics of the stream's substrate ( $D_{50}$  or average particle size) and the relative extent of impairment by sedimentation that is occurring ( $D_{84}$  or bankfull particle size).

#### ***QUALITY ASSURANCE/QUALITY CONTROL***

All individuals conducting field work will receive hands-on training prior to the field season. Individual training will occur simultaneously on the same stream so the results can be compared to the group average. Readings that deviate exceptionally from the norm will be examined for errors. In the field, Relative Bed Stability teams will consist of two people. Individuals who are more experienced in measuring Relative Bed Stability will be teamed up with the less experienced to assure reinforcement of training and accurate results.

## **SOP M. Forms used in the Watershed Assessment Section**

The forms used by the WAS are available internally via the WVDEP computer network. Navigate to G:\Wap\WASP\Forms\2005\WAP Habitat Forms

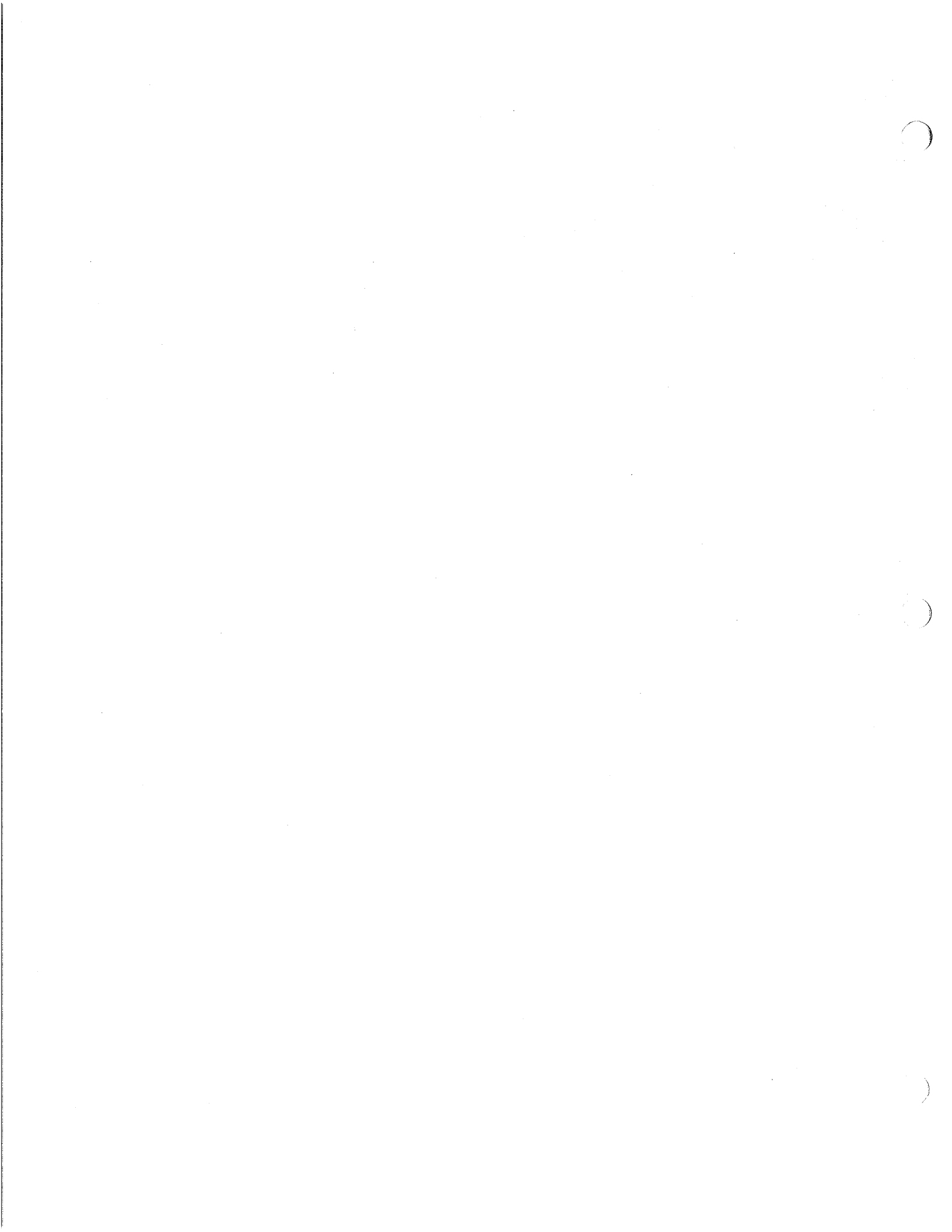




























<b>BENTHIC MACROINVERTEBRATE &amp; PERIPHYTON COLLECTION INFO&gt;&gt;&gt;&gt;&gt;&gt;&gt;</b>						Reviewers Initials		
ANcode			Date					
Benthic sample collected?		<input type="checkbox"/> Yes <input type="checkbox"/> No	If no, why?					
Benthic collection device		<input type="checkbox"/> Kicknet <input type="checkbox"/> D-net <input type="checkbox"/> Hand		Benthic habitat type & #		riffles		runs
Benthic sample comparability		Was benthic sample comparable with respect to riffle/run depth and velocity?					<input type="checkbox"/> Yes <input type="checkbox"/> No	
Is there evidence that the stream channel was scoured by recent flooding or high flows?							<input type="checkbox"/> Yes <input type="checkbox"/> No	
Is it possible that sample areas were dry or partially dry for an extended period before sample was taken?							<input type="checkbox"/> Yes <input type="checkbox"/> No	
Is there evidence that the stream is "wet-weather" and flowing only in response to recent rainfall?							<input type="checkbox"/> Yes <input type="checkbox"/> No	
Benthic kick area depths		m		m		m		m
Use the space below to describe the site and explain responses to the previous questions. What organisms were put in the jar?								
Inorganic Substrate (1 m <sup>2</sup> Of Kicked Substrate)		Class Codes		Size Class			% Composition	
Bedrock		BR		Smooth surface rock/hardpan (>4000 mm – bigger than a car)			%	
Boulder (BL)		BL		Basketball to car (>250-4000 mm)			%	
Cobble (CB)		CB		Tennisball to basketball (>64-250 mm)			%	
Coarse Gravel (CG)		CG		Marble to tennisball (>16-64 mm)			%	
Fine Gravel (FG)		FG		Ladybug to marble (>2-16 mm)			%	
Sand (SA)		SA		Gritty – up to ladybug (>0.06-2 mm)			%	
Silt & Fines (ST)		ST		Fine – not gritty (<0.06 mm)			%	
Clay (CL)		CL		Slick/ hard clay or hard-pan clay			%	
Enter estimated % composition for each substrate type. ****MACS SITES: estimate over entire 100 meter stream reach.****								
Describe quality of benthic substrate (i.e., stacked cobble and how it relates to the sample). Describe unusual substrate features and general comments about the site.								
Periphyton sample collected?		<input type="checkbox"/> Yes <input type="checkbox"/> No	If no, why?					
Periphyton Habitat & #		Riffle	Run	Shade and number of each		Fully exposed (0-25%)		Partly shaded (25-50%)
						Fully shaded (75-100%)		Partly exposed (50-75%)
Periphyton sample comparability		Was periphyton sample comparable? (Consider questions above about benthic comparability)					<input type="checkbox"/> Yes <input type="checkbox"/> No	
Use the space below to describe the sample and explain responses to the previous questions.								
Indicate abundance of each: 0=None, 1=Low, 2=Moderate, 3=High, 4=Extreme, NR=Not Rated		Periphyton (Brown-slick; Diatoms)		Filamentous Algae (green)		Aquatic Vascular Plants		"Aquatic" Mosses







**WV DEPARTMENT OF ENVIRONMENTAL PROTECTION - WATERSHED BRANCH**

Analysis Request Form, Rev. 12/05

TMDL WAP AWQN

Laboratory Name: \_\_\_\_\_ Circle Activity

Stream Name: \_\_\_\_\_ Watershed Name: \_\_\_\_\_

AN-Code: \_\_\_\_\_ Station Number: \_\_\_\_\_ Random #: \_\_\_\_\_ # of Containers \_\_\_\_\_

Sampled By: \_\_\_\_\_ Filtered By: \_\_\_\_\_

Sample Type: Water \_\_\_\_\_ Sediment \_\_\_\_\_ Other \_\_\_\_\_ Specified Method: **40 CFR 136**

Acid Lot #, Nitric: \_\_\_\_\_ Sulfuric: \_\_\_\_\_ HCl: \_\_\_\_\_ Field Meter # \_\_\_\_\_ Flow Meter # \_\_\_\_\_

Grab: Date-Time: \_\_\_\_ / \_\_\_\_ / \_\_\_\_ Lat: \_\_\_\_\_ Long: \_\_\_\_\_

Positioning Method (Circle One) Map GPS WCMS

Field Values<sup>1</sup>: Temp. \_\_\_\_\_ pH \_\_\_\_\_ D.O. \_\_\_\_\_ Cond. \_\_\_\_\_ Flow: \_\_\_\_\_

Pres.	Analysis	Pres.	Analysis	Pres.	Analysis	Tot*	Dis**	Preservation Code
3	Acidity (hot)	3	Tot. Solids	5	Sodium			1. None - Determined on-site 2. None 3. Iced 4. H <sub>2</sub> SO <sub>4</sub> to pH <2, Iced (Phenols in glass container) 5. HNO <sub>3</sub> to pH<2 6. (Cyanide) NaOH to pH>12, iced (0.6 g ascorbic acid used on samples with residual chlorine) 7. Sterile + 0.008% Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> , iced 8. Filtered immediately, iced 9. HCl to pH<2, iced 10. Other (Specify) REMARKS:  _____ # of Filters used *Indicate (Circle) desired detection level: H = Highte (e.g. 5 ppb) L = Low (e.g. 0.05 ppb) †Filtered in the field, nitric acid added
3	Alakinity	3	Dis. Solids	5	Aluminum	H	H L	
5	Hardness	3	Tot. Susp. Solids	5	Cadmium	L	L	
3	Sulfate	4	T. Phosphorus-P	5	Chromium	H L	H L	
3	Turbidity	3	Tot. Ortho PO <sub>4</sub> -P	5	Copper	L	L	
2	Chloride	8	Dis. Ortho PO <sub>4</sub> -P	5	Iron	H	H	
3	BOD5	4	TKN	5	Lead	L	L	
4	COD	4	Amonia-N	5	Manganese	H		
4	TOC	4	Unionized NH3	5	Mercury	H		
7	Fecal Coli., MF 24 hour holding	4	Org-N	5	Nickel	L	L	
		3	NO <sub>3</sub> -N (Nitrate)	5	Zinc	L	L	
7	Fecal Coli., MF 6 hour holding	3	NO <sub>2</sub> -N (Nitrite)	5	Calcium	H		
		4	Magnesium	5	Selenium	L		
7	E. Coli, Numeric	5	Potassium	5	Arsenic	L	L	
7	Fecal Strep.	5		5	Silver		L	
3	pH (lab)	3	Semi-Volatile					
3	Cond. (lab)		Organics					
3	Acidity (Cold)	9	Volatile					
			Organics					

Relinquished by:	Date & Time	Received By:	Relinquished by:	Date & Time	Received by:
		Lab:			Lab:

Mail Results to: ATTN: Janice Smithson (Lab Instructions: On invoice bill to Organization Unit 9480), WVDEP, WVDEP, DWWM, Watershed Branch, 601 57th Street, Charleston, WV 25304 Phone (304) 926-0499 ex. 1051, Fax. 926-0496

WHITE - Sample Collector Copy

CANARY -Laboratory Copy

**Benthic Sample – label for inside of jar**

Stream Name \_\_\_\_\_  
AN Code \_\_\_\_\_  
Date \_\_\_\_\_

**Benthic Sample – label for inside of jar**

Stream Name \_\_\_\_\_  
AN Code \_\_\_\_\_  
Date \_\_\_\_\_

**Benthic Sample – label for inside of jar**

Stream Name \_\_\_\_\_  
AN Code \_\_\_\_\_  
Date \_\_\_\_\_

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Stream Name \_\_\_\_\_  
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Date \_\_\_\_\_

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Date \_\_\_\_\_

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AN Code \_\_\_\_\_  
Date \_\_\_\_\_

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AN Code \_\_\_\_\_  
Date \_\_\_\_\_

**Benthic Sample – label for inside of jar**

Stream Name \_\_\_\_\_  
AN Code \_\_\_\_\_  
Date \_\_\_\_\_

**Benthic Sample – label for inside of jar**

Stream Name \_\_\_\_\_  
AN Code \_\_\_\_\_  
Date \_\_\_\_\_

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE

WV DEP WATERSHED ASSESSMENT PROGRAM MACROBENTHIC SAMPLE			
STREAM NAME			
ANCODE	RANDOM #	Circle one: Kick, D-net, MACS, or hand- nicked	
DATE	COUNTY	COLLECTOR	PRESERVATIVE







## WAP General Water Quality Form

						Reviewers Initials				
Stream Name (with location)										
ANcode					Date		Time		Geo	Bio
Basin			County			Quad				
GPS #	GPS St #		EPE		XY's Proofed			By		
Field Lat X-site				N	Field Lon X-site			W		
Corrected Lat				N	Corrected Lon			W		
Sampled	<input type="checkbox"/> Yes <input type="checkbox"/> No	If no, why?		<input type="checkbox"/> No Access-Physical Barrier ( <input type="checkbox"/> <i>Permanent</i> <input type="checkbox"/> <i>Temporary</i> ) <input type="checkbox"/> No Access-Landowner Denial ( <input type="checkbox"/> <i>Verbal Denial</i> <input type="checkbox"/> <i>Posted</i> <input type="checkbox"/> <i>Fenced</i> <i>Private</i> ) <input type="checkbox"/> Too Deep ( <input type="checkbox"/> <i>Permanent</i> / <input type="checkbox"/> <i>Temporary</i> ) <input type="checkbox"/> Dry <input type="checkbox"/> Filled <input type="checkbox"/> Impounded <input type="checkbox"/> Other:						
Sample Type		<input type="checkbox"/> YSI <input type="checkbox"/> Fecal <input type="checkbox"/> Nutrient <input type="checkbox"/> AMD <input type="checkbox"/> Flow <input type="checkbox"/> Other:				Duplicate Type		<input type="checkbox"/> None <input type="checkbox"/> Lab WQ Only		
Sample Location:		<input type="checkbox"/> Mid-Stream <input type="checkbox"/> Thalweg <input type="checkbox"/> Left Bank <input type="checkbox"/> Right Bank <input type="checkbox"/> Other:								
Hydrolab Method:		<input type="checkbox"/> Grab <input type="checkbox"/> Bucket <input type="checkbox"/> Other:			Lab Water Method: <input type="checkbox"/> Grab <input type="checkbox"/> Bucket <input type="checkbox"/> Other:					
Dup type:	<input type="checkbox"/> Lab <input type="checkbox"/> Fecal <input type="checkbox"/> Hab <input type="checkbox"/> Bugs <input type="checkbox"/> Periphyton			Dup number:		Was site moved (non-random)?		<input type="checkbox"/> Yes <input type="checkbox"/> No		
Explanation?										
Directions To Site										
<p>Sketch of Assessment Reach and Comments : Indicate North with (↑), indicate flow direction, indicate water sample (wq), indicate lat and long site with (X). Draw the sketch with a coarse resolution to give an overall idea of the sample area beyond the typical 100m reach.</p>										
Notes										

















































# Fish Survey Form

Sampling Date: \_\_\_\_\_ Time: \_\_\_\_\_ Stream/Lake Name: \_\_\_\_\_ ANCode: \_\_\_\_\_  
 Lat: \_\_\_\_\_ Lon: \_\_\_\_\_ County: \_\_\_\_\_ Topo Quad: \_\_\_\_\_  
 Collectors: \_\_\_\_\_ Collection Method(s): \_\_\_\_\_  
 Field Parameters: Temp. \_\_\_\_\_ C pH \_\_\_\_\_ D.O. \_\_\_\_\_ mg/l Cond. \_\_\_\_\_ umhos/cm Hydrolab/YSI#: \_\_\_\_\_  
 Circle Sample Type: Whole Fish Fillet Other: \_\_\_\_\_ Waterbody Type: \_\_\_\_\_ Water Collected? Yes No

Individual	Species	Length (cm)	Weight (g)	Composite # or Ind.
1				
2				
3				
4				
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6				
7				
8				
9				
10				
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Individual	Species	Length (cm)	Weight (g)	Composite # or Ind.
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**Filling out the Form:** A sheet (at least the front page) needs to be filled out for every site we visit or make a failed attempt to visit on a team's list, no matter if it was sampled or not. It does not matter what type of stream it was either, non-random, random, reference. This will help us track the streams more efficiently down the road. Location information should still be written down for these sites (e.g., topo name, coordinates, directions, etc.). If a site is moved any significant distance  $>0.5$ , then a form (front page) should be filled out for the original site stating why it was unsuitable & a form should be filled out for the new site making note of the original site.

**Non-random Sites - Where to Sample?** (also see Reference Sites below): Sample Target sites upstream of bridge or culvert, generally at the first riffle/run. Only sample a stream using MACS or Hand-Pick methodology if you are instructed to do so on the stream list. If a nasty discharge (AMD stream, point source outfall, etc.) falls within the chosen stream reach, slide the reach upstream of discharge & then take physicochemical samples upstream & downstream of discharge as well as from the discharge itself using a different form for each. If the discharge is a stream & is not on another crew's list, conduct a full WAP assessment on that stream. If you encounter this situation, document the situation thoroughly.

**Reference Sites:** Some potential reference sites have been identified & are marked as PRS on the crews stream list, these streams have few apparent problems, & should be sampled following reference site protocols. If the site marked on the map is not suitable, try scouting upstream to locate a better site. Every effort should be made to sample the stream as far down in the watershed as allowed by disturbances. Other potential sites have not been identified & are not marked 'PRS' on the crew's list. It is the crew's responsibility to determine if these sites will meet our criteria (see Reference Site section of SOP) for reference sites. Unless otherwise directed to sample them (e.g., via a note on the stream list), do not sample these sites if there are obvious impacts at these sites. If they do look good, then sample them as reference sites. If a potential reference site is encountered that is not listed on the stream list & you are sure no one has it on another list, then sample it if it meets the requirements. **Very important:** If possible, recon the watershed above the PRS & make notes about the land use quality.

**Locating the Random Site:** USGS topographic maps with a 1:24,000 scale are marked with an **X (highlighted in pink)** to designate the sampling station for random sites. This spot is referred to as the **X-site** & is the **downstream** end of the 100 m reach that is to be assessed. *Note: In the past, the X-site was the center of the 100 m reach.* If you have confidently navigated to the X site, check GPS readings to make sure you are within 10 seconds of the X-site latitude & longitude. If you have good satellite coverage on your GPS (i.e., low EPE), move as close to the provided coordinates as possible. Crew members should collaborate & utilize best professional judgment (BPJ) to decide where the X-site is located (finely tuned map reading skills are important). If GPS doesn't work, do the best you can using the topo & indicate so on the field assessment form.

**Determining Random Site Status:** Benthic macroinvertebrate data is the primary tool driving the random program. If a benthic macroinvertebrate sample cannot be collected at a site, skip it & move on to the next site. Always complete the first page of the field assessment form regardless of whether the site is sampled or not. All unassessed (as well as assessed) sites will be included in the final analysis of the data. So, it is very important to thoroughly document the reason why a site was not sampled. After establishing the X-site, thoroughly examine the assessment reach & determine if it fits in our target population of random sites. Our target population consists of the following: **1) 1<sup>st</sup> thru 4<sup>th</sup> order streams (check stream list provided by Janice), 2) riffle/run habitat, 3) wadeable, 4) can be sampled using kick (rectangular frame kicknet or D-net) protocols that result in comparable data.** Don't confuse riffle/run streams that are heavily embedded (sandy/silty) with true "wetland-like" streams. If stream velocity will effectively carry organisms into the net, then do your best to collect a sample. Look for contours (if present) in the sand/silt & collect sample there. Also, **do not** avoid sites that are all bedrock or have large substrates (boulder). In the final data analysis, these sites can be classified as less productive habitats & not necessarily impaired. Sample bedrock at areas where there are contours, fissures, &/or rough places (if present), etc. Sites to avoid include the following: 1) too deep to wade safely, 2) too deep to get a comparable benthic sample (water flows over the net) 3) too shallow &/or slow to get a comparable sample using our kicking techniques.

**Sliding the Random Assessment Reach:** **Important Note: The X-site is the downstream end of the 100m reach.** In some instances, it may be necessary to **slide** the reach. Remember: never move the X-site, just slide the reach around it. Do not proceed upstream into a lower order stream when establishing the 100 m reach. If such a confluence is encountered, note the distance & mark the confluence as the reach end. Make up for the loss of the reach length by sliding the other end of the reach an equivalent distance away from the X-site. **NOTE: the confluence must be within the initial 100m reach for this sliding to apply.** Additionally, if the reach contains a lake, reservoir, or pond, mark the water body as the reach end & make up for the loss of the reach length by moving the other end of the reach an equivalent distance from the X-site. However, if the X-site is completely within a lake, pond, or other such impoundment, no sampling can occur & only the front page of the habitat form is to be filled out describing the situation. Also, don't slide the reach downstream into a larger order stream. This may occur when you are sliding the reach downstream to avoid lakes, ponds, etc. **Unlike previous years, do not slide the reach to encompass better habitat or to get more riffle/run habitat to sample.** Also, do not slide the reach to avoid man-made obstacles such as bridges, culverts, rip-rap, or channelization.

**Water Quality Parameters:** *Take Hydrolab readings & Fecal coliform at every site!*

**Random & Potential Reference Sites:**

- Acidity (Hot), Alkalinity, Sulfate, Chloride, Fecal coli., TSS, Tot. Phos., TKN, NO<sub>2</sub>-NO<sub>3</sub>-N, Mg, Al (Tot. & Dis.), Cu (Dis.), Fe (Tot. & Dis.), Mn, Hg (0.1 ug/L MDL), Zn (Dis.), Ca, Se (Tot. & Dis.). (Note: Order Low Level Detection on Tot. & Dis. Cu, Zn, Se.)  
**4 cubies (iced, HNO<sub>3</sub>, filtered HNO<sub>3</sub>, & H<sub>2</sub>SO<sub>4</sub>) & fecal**

**AMD Parameters:** Take when: 1) **conductivity alone is > 500**, 2) pH <6.0 & conductivity is >200, 3) if stream is on the 303(d) list for AMD, or 4) if for any reason you suspect mine drainage:

- Acidity (Hot), Alkalinity, Sulfate, Chloride, Fecal coli., TSS, Al (Tot. & Dis.), Fe (Tot Only), & Mn. Take Ammonia-N (NH<sub>3</sub>) if it is suspected that Ammonia is being used to treat the stream water. **3 cubies (iced, HNO<sub>3</sub>, & filtered HNO<sub>3</sub>) & fecal**

**Acid Rain Parameters:** Take when: 1) pH <6.0 & conductivity is <50, 2) if stream is on the 303(d) list for pH unrelated to mining, or 2) if for any reason you suspect acid rain deposition impacting the stream:

- Acidity (Hot), Alkalinity, Sulfate, Fecal coli., Acidity (Cold), TSS, Al (Tot. & Dis.), Fe (Tot Only), Mn, & Ca (Tot.).  
**3 cubies (iced, HNO<sub>3</sub>, & filtered HNO<sub>3</sub>) & fecal**

**Nutrient Enrichment:** Take within 24 hours of a significant rain or when animal waste, straight pipes, STP outfalls, etc., may be impacting the stream:

- TSS, Tot. Phos., TKN, NO<sub>2</sub>-NO<sub>3</sub>-N, & Fecal coli. Take Ammonia-N (NH<sub>3</sub>) if cattle or other livestock have direct access to stream or if there is evidence of possible ammonia input.  
**2 cubies (iced, H<sub>2</sub>SO<sub>4</sub>) & fecal**

**Oil & Gas:** Take if oil or gas activities are evident & cond. >200 in absence of other sources like AMD:

- Chloride & Fecal coli.  
**1 cubie (iced) & fecal**

**Other Water Quality Notes:**

- Place fecal bottles in separate zip-lock sandwich size baggies before putting in ice (do not submerge in ice water!).
- Label each container w/ WVDEP WAP, stream name, AN-code, date/time collected, collector (especially if a dup.), & preservative type.
- Take water samples at lower end of reach for Non-Random targeted sites. **Take water samples at lower end of 100 m assessment reach for random sites regardless of the location of the X-site.**
- **If Alkalinity is being analyzed, 100% of the air must be expunged from the unfixed cubitainer to avoid contamination.**
- **Remember:** A net minimum of 200 mL of filtered sample should be turned in for dissolved metal analysis at **most labs we deal with.**

**Flow:** Locate the best cross section to measure. (Flow parallel to stream bank, minimum of large disturbances, no large eddies, flow not heavily skewed to one side, etc.). Move some substrate to create a spot

- Determine the wetted width & effective flow (E.F.) width of the stream. Record distance from bank to edge of E.F.
- Divide the effective flow width into at least 20 segments for streams **over 10ft width** or into at least 10 segments for streams > 3ft or 1m. If smaller than 3ft, you should use fewer segments & best professional judgment.
- Take the first measurement at the point that is approximately one half the determined segment length from edge of E.F.
- Velocity is measured at 6/10ths of depth at each transect (determined for you if using normal wading rod). If the depth is greater than 2 feet, take two velocity readings at that point: one at 0.2 depth & one at 0.8 depth.
- Record segment distance & depth in tenths of feet & velocity in ft/second.
- Any negative flow readings should be recorded as zero with a note that a negative value was observed.
- Continue taking measurements along the transect by tightening the increments in plumes of high velocity or radically changing depths or by spreading them out in relatively consistent velocity/depth zones until the last measurement is around one half the average segment length from the other edge of E.F. **The minimum width increment that can be used is 0.3 ft or the approximate width of the probe head** as any increments smaller than this will be indistinguishable from one another.
- If caught behind an unmovable boulder or other obstacle, you need to take the measurement above or behind the boulder & also on either side of that boulder to get data from higher velocity plumes deflected around the boulder.
- **Remember: The flow measurement probe is not accurate in depths less than 0.2 ft as it makes the flow measurement unreliable. Consider this carefully when choosing a location for flow measurement. If a stream simply does not have areas with at least 0.2ft depth, then take what measurement you can & indicate that the flow is not comparable on the form & why. Always not flow comparability & unusual situations.**
- Store the flow meter head separate from the rod to avoid damage & keep the physical structure of the head clear, consistent & smooth.

**Note:** Extreme conductivity could have an effect adverse on the flow reading; be aware & on the lookout for this when working in such streams.

**Field Blanks:** Each team should prepare field blanks at the sites designated by Janice at the time the normal samples are collected. Distilled, deionized water is the blank "sample". Crews should "collect" well-sealed container(s) of DI water (ran from the Nano-pure filter) at the beginning of the week. When the crew arrives at a designated site, they should prepare an exact set of cubitainers & preserve the blanks as they would normal samples (this includes filtering one blank for dissolved metals). Use one cubitainer for each type of preserved sample collected (or expected to be collected) that week. **Do not prepare a field blank for analysis that requires unfixed samples.** Blanks are labeled with a false AN-code & stream name (e.g., Surbaugh Branch, KP-100-A-B). A separate "Analysis Request Form" is completed for the field blanks with faked hydrolab data on the form. Do not indicate on any paperwork given to the lab that it is a field blank until after the sample is submitted to the lab.

**Duplicate Sites are not Optional !!** Attempt to do the duplicate site at the designated site. If the first site you visit on a list provides enough good habitat to do a duplicate, then sample it as a duplicate. Do not wait until the end of a week or list to sample for a duplicate stream. Duplicate sites should be large enough to obtain two complete benthic samples (4 kicks versus 4 kicks) in comparable habitat (substrate size & velocity in particular), if not, select another stream. Make sure the in stream substrate & velocity of the benthic sampling sites are as similar as possible. Make sure to write the name of the collector & Dup #1 & Dup #2 on the appropriate sample containers. Each team member should complete the habitat forms & collect macroinvertebrate & appropriate water samples as if they are the only person there (i.e., a one person crew). GPS values can be shared. Hydrolab & flow values should only be recorded on the Dup #1 form. Only filter your sample!

**Biology:** *Check the nets for holes regularly & empty after every kick; always use the small brush on cobble & larger rocks. Put a label on the inside & outside of every sample jar. Sample a variety of riffles (high gradient, intermediate, & low gradient or flat), but sample the most productive or best ones within each group available.*

**Comparable Methods:**

- Streams w/ riffles & of adequate size, use the **Modified Kick (Surber) net = 4 kicks - 0.5m x 0.5m, 20 seconds each.**
- Streams w/ riffles but too narrow to use Surber, use **D-net = 9 kicks - 0.3m (1 foot) x 0.3m, 20 seconds each.**

**Non-comparable methods (use only if directed to do so on the stream list):**

- Streams w/ too low of a flow to use other protocols, conduct a **Hand pick = 4 areas (0.5m x 0.5m) thoroughly** in wetted connections between pools, not in pools. It may not be possible to sample a square 0.5m x 0.5m area (stream too narrow), it is possible to sample non-square areas like 0.25m (~10 inches) x 1.0m. Remember that hand pick samples are not comparable, but this type of sampling can still provide valuable data in certain situations.
- Streams w/ no flowing water - use **Multi-habitat (MACS) = 20 jab/sweeps - 0.5m each.** Sample the habitats in proportion to the productive habitat in the reach (i.e., if 50 % of the productive habitat is woody snags, sample 10 woody snags). **Do MACS sites only if absolutely necessary & if there is plenty of proper habitats to sample.** Remember that MACS samples are not comparable, but this type of sampling can still provide valuable data in certain situations.

**Sample Preservation** – The samples must be properly preserved!! Each sample must have at least 70% ethanol to adequately preserve organisms. If the sample jar is very full (especially with lots of organic material like filamentous algae or leaves), divide sample into two or more labeled jars. Each sample jar should be no more than half full with the benthic sample. **Be sure to clearly indicate '1 of 2' & '2 of 2', etc. on the jars.** Also, tape any multiple jar samples together back in the lab. **Do not invert the sample jar after putting the organic material (Bugs & Leaf Bits) in on top of the inorganic material (Sand & Silt).**

**Periphyton:** *Do at all Random, Reference, TMDL Bio sites & those Non-Random sites with nutrient enrichment or as directed on the stream list. Periphyton may be collected at big streams that are too deep for benthos collection (i.e., water over the net) but not too deep to reach in and grab cobble to sample periphyton as a substitute for the benthic sample.*

In order to make our collections as consistent as possible, collect periphyton from the same riffle & run habitats of the stream as the benthic macroinvertebrates are collected. Collect five cobble-sized rocks that are exposed to light (if possible) from throughout the reach. Record the number of rocks sampled from riffles & from runs on the field form. Also, record the number of rocks collected in each light/shade category. Using the PVC ring to delimit the sample area, use the spatula to scrape **all algae** from the **upper surface** of the rocks into the sample jar. Use a toothbrush to loosen any remaining periphyton. Remove the sampler & rinse loosened algae into the sample jar. Periphyton from the five rocks is composited into one sample jar. If there are no rocks available from the reach, collect periphyton from removable wood (same technique as for rocks), documenting on the field sheet exactly what was sampled. Rinse the sampler & brush &/or scraping device thoroughly with stream water to avoid contamination of subsequent collections before and after each sample event. Assuming the sample jar is about 3/4 full (about 120 mL), a guideline is to preserve with a 5mL (100 drops) of formalin for sparse to normal periphyton amounts; add more for samples with heavy amounts of green algae. Take extra care when preserving, as formalin is a known (suspected?) carcinogen. **Note: Samples do not need to be preserved with formalin immediately.** It is probably easier to preserve all periphyton samples collected in a given day at one time-upon returning to office or hotel parking lot. Whether samples are fixed immediately or not, **they should be placed in a cooler with ice.** Sample jars should be taped to minimize leaking & should have a label on the inside & outside.

**Relative Bed Stability:** *Do at all the first two Random sites on the stream list if possible (or the next available site) & those Non-Random sites as directed on the stream list. All measurements should be recorded in meters.*

**A) Pebble Count:** Establish the sample reach for pebble counts by multiplying the average stream width (taken at d.s., u.s., & mid. of the 100m reach) by 40 (e.g., if the average width equals 6 m, the pebble count reach would be  $240\text{ m} = 6\text{ m} \times 40$ ). The reach minimum is 100m & the reach maximum is 500m. After the reach length is established, divide by 10 to get the distance between transects (A thru K=11 transects). For example, if the reach length equals 240 m, transects would be established 24 m apart ( $240/10=24$ ). Starting at transect A (the downstream end of the original 100m reach), measure the wetted width of the stream with the surveyor pole (or tape). Divide the width by 4 to get the left, left-middle, middle, right-middle, & right measurement points. One bank measurement is always taken at zero & the opposite bank measurement is taken at the value that equals the stream width. The thalweg pole is used at each measurement point to determine the substrate particle type for that point. The substrate particle that the point of the thalweg pole falls on is measured, classified, & recorded based on the substrate classification table on the field form. Determine the thalweg by measuring the deepest point of the stream along each transect. Read the depth on the side of the thalweg pole to avoid the wave produced by the pole in the moving water. These measurements continue from transect A through transect K (uppermost transect). Be sure the data from each transect is recorded in the appropriate space on the field form. **Note for if you encounter split channels: If the split channel features a bar (*bar definition: a channel feature below the bankfull height that is dry during baseflow conditions*) then conduct the measurements at that transect as if there was only one channel and note the presence of the bar. Any measurements that fall on the bar should be treated just as if it was inundated with water, but noted as being taken on a bar. If the split channel features an island (*island definition: a channel feature even with the surrounding flood plain or above the bankfull height that remains dry even at bankfull flow*) then conduct a separate transect in each channel for the length of the island. The situation should be documented and the second transect information is recorded continuing the transect letters down the alphabet starting with J. Be sure to document either of these or extraordinary cases well on the field form.**

**B) Bankfull Height:** Defined as the channel height that is filled by moderate-sized flood events that occur every one or two years. Bankfull height can be determined by looking for an obvious slope break that differentiates the channel from a flat floodplain higher than the channel. A transition sometimes exists between exposed substrate & vegetation, which can also mark the bankfull height. Also, it may be determined by moss growing on rocks along the banks. Deposited drift materials (leaves, sticks, trash) along the bank can sometimes mark the bankfull height. Record a **minimum of three bankfull height measurements** throughout the reach as you conduct the pebble count (but still try to get as many as possible).

**C) Gradient:** For Random Sites, the measurements start at the upstream end of the reach (as previously defined by the pebble count method) & will move to the downstream end. **Do at all Random Sites for 100-500m variable reach as a part of the Relative Bed Stability**

**Handheld Eye Level Method** - Slope is measured by "backsiting" or "backshooting" downstream between the two reach ends. The primary method is to use a handheld eye level. If a situation occurs where using the handheld eye level is not feasible (e.g., the stream is too sinuous or there is too much overhanging vegetation) then the water level may be used instead to cover that distance.

**Note:** *Each individual should determine and remember their eye level height (the point on the survey pole that their eye is level with) before doing any slope measurements. Also, keep in mind that this point can change when wearing different wading boots or footwear.*

- 1) One individual stands at the water surface along the bank with the handheld eye level while the other holds the survey extension pole at the water surface downstream (as far as the individual can see accurately with the handheld eye level).
- 2) Looking through the eye level, the upstream individual determines where along the vertical surface of the survey extension pole their eye is level. The upstream individual will instruct the downstream individual to move a horizontal marker (e.g., finger, stick, pencil, thalweg pole) up or down to the same spot. The upstream individual's eye level height is then subtracted from the measured height to determine the rise of the stream for that distance. This value is recorded on the field form.
- 3) The upstream individual must now move to the position of the downstream individual (which can be marked by stacking rocks or with placement of an object or flagging) and the downstream individual moves down as far as the upstream individual can see accurately with the handheld eye level). Repeat the measurements as described until the downstream end has been reached.

**Important:** Gradient measurements must be taken along the full length of the reach. If the full reach is not measured with the tape measure, the gradient calculation will be incorrect & the time taken to record this data will be wasted.

**Notes** - If you encounter a high waterfall in the reach, measure the rise from the edge (if safe) of the fall to the splash-zone below using the surveyor pole & record it in one of the extra blanks on the field form. Also describe the reading & include what it is (i.e. waterfall) & where in the reach the waterfall was located (transect location). Then continue measurements past the waterfall as normal.

- If a situation occurs where using the handheld eye level is not feasible (e.g., the stream is too sinuous or there is too much overhanging vegetation) then the **Water Level Method** may be used instead to cover that distance as outlined in SOP L.

Current Water Quality Standards

Parameter	Water Temperature Regime		Water Quality Standards		Units		
	Cold or Trout Water		Deg F				
	Warm or Warm Water Fishery		>70	>87			
Use Designation	B1 - Warm Water Fishery, B4 - Wetlands		B2 - Trout		C - Contact Recreation	A - Public Water Supply	D - Agr/Wildlife, E - Industrial Water Supply
	acute	chronic	acute	chronic			
Bio Impairment	<=60.6	<=60.6	<=60.6	<=60.6			
Fecal Coliform, geo mean- 5 samples min/more than 10% of samples, per 100 ml					200/400	200/400	
Fecal Coliform, per 100 ml, Ohio R. November - April, geo mean 5 samples min.					2000		
DO, any time, mg/l minimum	<5	<5	<6, <7 when spawning	<6, <7 when spawning	<5	<5	<5
DO, any time, mg/l minimum (Ohio R; Kan. Zone 1)	<4	<4					
DO, any time, mg/l minimum (Ohio Apr. 15 - June 15)	<5	<5					
DO, daily avg, mg/l minimum (Ohio R)	<5	<5					
pH	<6.0 or >9.0	<6.0 or >9.0	<6.0 or >9.0	<6.0 or >9.0	<6.0 or >9.0	<6.0 or >9.0	<6.0 or >9.0
Aluminum (D), mg/l	0.75	0.087	0.75	0.087			
Iron (T), mg/l		1.5		0.5		1.5	
Manganese (T), mg/l						1	
Arsenic (T), mg/l					0.05	0.05	0.1
Cyanide (as free), mg/l	0.022	0.005	0.022	0.005	0.005	0.005	
Mercury (T), mg/l	0.0024		0.0024		0.00015	0.00014	
Selenium (T), mg/l	0.02	0.005	0.02	0.005		0.01	
Chromium hexavalent (+6), diss, mg/l	0.01571	0.01058	0.01571	0.00693			
Ammonia (Un-ionized) mg/l	Need separate spreadsheet						
Chloride, mg/l	860	230	860	230	250	250	
Nitrate (as Nitrate-N), mg/l						10	
Nitrite (as Nitrite-N), mg/l	1		0.06				Hardness Dep
Cadmium (D), mg/l	Hardness Dependent						
Lead (D), mg/l	Hardness Dependent						
Nickel (D), mg/l	Hardness Dependent						
Silver (D), mg/l	Hardness Dependent						
Zinc (D), mg/l	Hardness Dependent						



Parameter	B1 - Warm Water Fishery, B4 - Wetlands		B2 - Trout		C - Contact Recreation	A - Public Water Supply	D - Agr/Wildlife, E - Industrial Water Supply
	acute	chronic	acute	chronic			
Antimony (T), mg/l					4.3	0.014	
Arsenic (D) Trivalent, mg/l	0.36	0.19	0.36	0.19			
Barium (T), mg/l						1	
Beryllium(T), mg/l	0.13		0.13			0.0000077	
Cadmium (D), mg/l (Ohio R)						0.01	
Chromium trivalent (+3), diss, mg/l			Hardness Dependent				
Copper (T), mg/l						1	
Copper (D), mg/l			Hardness Dependent				
Fluoride (T), mg/l						1.4	2.0 (for D)
Lead (T), mg/l						0.05	
Methylmercury, mg/l			0.000012				
Nickel (T), mg/l					4.6	0.51	
Silver (T), mg/l			Hardness Dependent				
Thallium, (T) mg/l					0.0063	0.0017	
Threshold Odor					>8 at 104 deg. F	>8 at 104 deg. F	
Total Residual Chlorine, mg/l	0.019	0.011	No chlorinated discharges				
Turbidity		X	X		X	X	

**Fish Tissue Standard**

Mercury in Fish	0.5	Body Burden value, ug/g

**Some Basic Flag Codes for the Field Sheets**

<b>\$</b>	See another sample record for the same data (e.g., Duplicate Sample)
<b>I</b>	Parameter not recorded or deleted due to instrument problems or maintenance issues
<b>L</b>	Parameter recorded but suspected to be incorrect value; There is a low probability that the value is incorrect
<b>M</b>	There is a moderate probability that the value is incorrect
<b>H</b>	There is a high probability that the value is incorrect

**ADB Flag Values**

Conductivity	500	umohs
<b>Metals:</b>		
Total Aluminum	0.75	mg/L
Sulfate	170	mg/L
Hardness	200	20-75 soft
Chloride	230	mg/L
Nitrate plus Nitrite	2	mg/L
Phosphorus	0.1	mg/L
Total Suspended Solids	15	mg/L

## 2005 Field Equipment Checklist

<b>Personnel Equipment</b>		
	Waders	Essential...our signature piece of equipment
	Rain Gear	Stay Dry...or mostly Dry
	Personal Gear	Luggage, etc.
	Water Bottle	Stay Hydrated!
	Personal First Aid Kit	Small Red Fanny Bags
<b>Weekly Needs</b>		
	GPS Units	Garmin and/or Trimble (Keep in Camera Bag or Backpack)
	YSI or Hydrolab	Take inside at end of week; Calibrate at beginning of week or day.
	Camera	Sony or Kodak (Take inside at end of week)
	Ice	From Ice Machine in the general garage area or Store
	95% EtOH	In Water's Garage Area Temporarily; Soon will be in Cylinder Storage
<b>Paperwork and Clipboard Stuff</b>		
	Stream List	Get Assignment from Janice
	Maps	Topographic, WV Gazetteer, WV County Roads
	Habitat Forms	Main WAP Form, Write-In the Rain Version, Front Page only version (for Dry or Access denied sites), Flow Appendix Sheet, Glide/Pool RBP Appendix Sheet, TMDL Forms; all on the G: drive
	Analysis Request Form	In the Water Lab; Extras in Karen Maes's Office
	Bug Jar Labels	Inner (on waterproof paper) and Outer; on the G: drive
	Periphyton Labels	Same label for inside and out; on the G: drive
	SOP	Get a copy from Janice or the G: drive
	Personal Field Books/Notepads	For the biomorph to write notes on while the Geomorph is away. Extras in Water Lab
	WAP Brochures/Pamphlets	To give to interested parties while in the field
	Data Request Forms	To give to interested parties while in the field; Get from Karen Light
	Clipboards	Assigned to each person
	Pencils and Pens	Get out of our Cabinet in the Mail Room or See Karen Light for Office Supplies
	Compass	To get your North Bearing
	Laptop	Update w/ Maps, WCMS Projects, Shapefiles, and Database as needed
<b>Jeep Maintenance</b>		
	Fix-A-Flat	Action Packer or under backseat.
	Come-Along	Often stored under backseat.
	Tow Straps	Often stored under backseat.
	Collapsible Shovel	Often stored under backseat.
	Axe	Often stored under backseat.
	Tools	Screwdrivers, wrenches, ratchets, etc.
	Flashlight	In Action Packer?
	Car Phone	Hide in vehicle in console or glove box.
	Lab & Vehicle Phone Number Sheet	Should always stay in vehicle in glove box, middle console, or under sun visor.



## 2005 Field Equipment Checklist

<b>Miscellaneous Jeep Stuff (Safety and Backup Supplies)</b>		
Action Packer	To store seldom used equipment	
First Aid Kit	Should be checked regularly	
Fire Extinguisher	Should be checked regularly	
Latex Gloves	To protect hands in nasty looking streams; Refills kept in Water Lab	
Handi-Wipes	To approximate cleaning of the hands. Refills in Water Lab	
Insect Repellant	DEET works the best; spray clothing only and keep off skin and out of eyes and mouth	
Blaze Orange Vest	Extras in Water Lab	
Life Vest/PFD		
Pepper Spray		
Machete		
Toilet Paper	"Acquire" from bathroom or hotel ☺	
AA Batteries	For GPS and Hydrolab Units; New ones in Water Lab	
C Cell Batteries	For YSI's; New ones in Water Lab	
D cell Batteries	For Flow Meter; New ones in Water Lab	
pH Standards	To recalibrate or check pH probe in field; In Water Lab	
Conductivity Standards	To recalibrate or check Conductivity probe in field; In Water Lab	
DO Maintenance Kits	To replace a DO membrane in field; Kept with YSI/Hydrolab; Extras In Water Lab	
pH Strips	To test sample fixation	
<b>Water Quality Sampling (TMDL and WAP)</b>		
Fecal Coliform Bottles	In Water's Garage Area	
Cubitainers	In Water's Garage Area	
Lids for Cubitainers	Used if Cubitainers do not come with lids; In Water's Garage Area	
HNO <sub>3</sub>	Refills in Water Lab; One extra in Action packer?	
H <sub>2</sub> SO <sub>4</sub>	Refills in Water Lab; One extra in Action packer?	
New Filter Apparatus (Primary)	Drill (w/Rechargeable Batteries and Recharger), Peristaltic Pump Board, Tubing, Disc and/or Cartridge Filters; Replacement Tubing and Filters are in the Water Lab	
Old Filter Apparatus (as a backup)	Funnel, Filter Sieve, Flask, Hand Pump, Filter Papers, DI Bottle; Acid wash once at end of week and store in large Zip-Lock; Old extra ones in Water Lab; Make sure DI Bottle is not contaminated-Wash regularly	
Plastic File Case	To keep the filter apparatus or supplies clean and dry	
Sharpie	New ones in Water Lab	
Small Zip-Lock Bags	To store individual fecal samples; Refills in Water Lab; One extra in Action packer?	
Large Zip-Lock Bags	To store multiple fecal samples; Refills in Water Lab; One extra in Action packer?	
Paper towels	To dab away excess water; steal from janitor's closet.	
Stainless Steel Bucket	To sample from bridges	
Rope	To sample from bridges; Also part of Jeep Maintenance	
Large Ice Chest	To store samples in field; In Water's Garage Area	
Field Blanks (DI Water)	Get from USGS jugs (temporary) or Still (future) and mark with a check	

## 2005 Field Equipment Checklist

<b>Flow Measurement (TMDL and WAP)</b>		
	Measuring Tape	Marked in tenths of a foot; Do not use metric; New ones in Water Lab
	Flow Rod w/Vector Ribbon	Marked in tenths of a foot; Do not use metric.
	Flow Meter	Numbered per unit
<b>Photography (TMDL and WAP)</b>		
	Extra CD Disks	3"CD-R/RWs or Memory Sticks depending on model
<b>Macroinvertebrate Sampling (Mainly Summer WAP)</b>		
	Surber-on-a-Stick	Remember to check for holes
	D-Net	Remember to check for holes
	Brush	To scrape at cobble and boulders; One extra in Action Packer would be a good idea
	Forceps	To pick macroinvertebrates off nets; One extra in Action packer?
	Sieve	To sift through benthic material; Extras in Water Lab; One extra in Action packer?
	Wash Bottle	To rinse benthic material; doesn't need to be clean
	Plastic Bucket	To temporarily store benthic material
	Bug Jars	Large sizes for more permanent storage; In Storage Area at BC
	Old Ice Chest or Box	To store Bug Jars
	Clear Packing Tape	Refills in Storage Cabinet in Water Lab
	Tape Dispenser	Refills in Storage Cabinet in Water Lab
	Macroinvertebrate Book	Keep in the jeep for field bug reference
<b>Periphyton Sampling</b>		
	Scraping Tool	To scrape at the rocks; A microspatula or spoon-type instrument; Extras in in Water Lab; One extra in Action Packer?
	Small Brush (Toothbrush)	To brush at the rocks; One extra in Action Packer? Change once a week.
	Sample Container	4 oz "specimen jar"; In Water Lab
	10 % Formalin	In a labeled dropper style bottle: WATCH YOUR EYES! Stored under sink in Water Lab
	Electrical Tape	To seal the periphyton container; Refills in in Water Lab
	Inner Tube Sampler	
	Small Ice Chest	To separate periphyton samples (nasty formalin) from the other water samples.
<b>Sediment Measurement (Random Sampling Only)</b>		
	100 m Measuring Tape	Extras stored in Water Lab
	Thalwag Pole	
	Surveyor Pole	
	Handheld Eye Level	
	Water Level Tubing	Use as an alternate to the Handheld Eye Level
	2 Clamps or Velcro straps	
	Flagging	To mark Transects and sites for easy identification; Extras in Water Lab

## SOP N. SOURCE SAMPLING PROCEDURES FOR TMDL MONITORING

**AML:** Some AML sources are selected to be included on the TMDL sample lists and will be sampled quarterly for AMD parameters and flow. One may encounter other AML sources on these and other streams that are not on this sample list while sampling. A source form can be filled out for these other sources as it is deemed appropriate. Include at least field readings, estimated flow, GPS coordinates, pictures, and the visible impact on stream. A sample is not needed unless it is suspected that this is a one-time event or intermittent source. The Field Supervisor will forward a copy to the Source Tracker and they will let you know if any follow-up is needed. Generally, the Source Tracker will follow up with additional source sampling.

**Permitted Sources:** If it is suspected that a permitted discharge is not within permit limits and is having a negative impact on the stream sample on a particular day, it would be very helpful to the TMDL model to have a sample of the permitted discharge (and its flow) on the same day that the stream sample is taken.

**Other Sources:** It is not necessary to collect samples from "common occurrence" pollutant sources like pastures or log jobs. Reserve source sampling for the rare or severe instances that are impacting the stream samples on a particular day. If it is suspected that runoff from a specific source (*e.g.*, problem log job or dairy manure pond overflow) is impacting the stream sample, go ahead and get a source sample if it is practical to do so. If it is not practical to get a sample, any documentation that can be provided will be helpful (*e.g.*, GPS coordinates, directions, pictures). Any general source information that may need to be passed on to the Source Tracker can be sent via email.

## Appendix C

### Watershed Branch Analytical Contracts

C-1 Water Analysis

C-2 Macroinvertebrate Identification

C-3 Periphyton Identification

Appendix C-1

Contract Specifications

For

Water Quality Analyses

## SPECIFICATIONS

### Releases:

In addition to consideration for vendor location and workload, releases or work orders shall be issued to the vendor with the estimated lowest cost to the state, as determined by DEP. Historically, the following West Virginia laboratories have provided inorganic laboratory services to the Division of Water Resources (DWR) and the Environmental Enforcement Unit (EE), and are considered qualified vendors and who submit bona fide bids.

Analabs Inc, Beckley

Bio-Chem. Testing -Teays, WV

CT &E -Charleston, WV

N.E.S.T. -Belington, WV

-- Hydrochem -Shenandoah Junction, WV

REIC -Beaver, WV

Sturm -Anmoore, WV

Tra-Det -Triadelphia, WV

R.D. Zande & Associates, Buckhannon, WV

Reliance Laboratories, Bridgeport, WV

The Department of Environmental Protection's (DEP) Division of Water Resources (DWR) and the Environmental Enforcement Unit (EE) conducts inspections of permitted and non-permitted facilities, investigates complaints, monitors ambient quality of surface water, groundwater and sediments, performs studies, and provides water quality information to the citizens of West Virginia and other government agencies. Legal action based upon analytic results is possible. Therefore, the firm or firms selected must have a quality control program in place and meet the following qualifications:

1. A laboratory, preferably, but not necessarily, located in West Virginia for the maximum utilization by DWR and EE personnel.
2. Chemist on Staff experienced in water analysis and its interpretation.
3. A full time Quality Assurance / Quality Control officer on staff experienced in water quality analysis.
4. The laboratory must be certified by the Water Resources Quality Assurance Program.
5. Be accessible by telephone 24 hours per day, 7 days per week. (Answering machine not acceptable)
6. Capable of attending and providing expert testimony in legal proceedings, upon request.
7. Have the ability to analyze for all parameters contained in at least two categories, (exclusive of category 5) in the Bid Package.

## SCOPE

In administering and enforcing most of the pollution control laws of the state, the importance of quality control cannot be overstated. Quality control measures must be strictly adhered to in all phases of sample collection, preservation, transportation, and analysis. The quality control and analytical work, as they relate to the contractor's responsibility, is divided into four (4) major steps:

STEP 1 -Collection of sample from specified office.

STEP 2 -Conduct specified analysis on samples in a timely and professional manner.

STEP 3 -Establishment of continuing program to ensure the reliability of analytical data.

STEP 4 -Legal Testimony

### Step 1 -Collection of Samples from Specified Locations

The sampling for the DWR and EE shall be conducted by Division personnel. The vendor shall be notified of the date sampling occurs or is to occur and from which OWR and EE office or other location the sample can be obtained. The vendor shall be notified when the sample was taken (time/date) and the person who collected the sample. The vendor shall be responsible for obtaining the sample from the specified location and delivery of sample to the laboratory within 24 hours from the time of sampling. The vendor shall indicate the time the sample was obtained from the pickup location and its condition and the time the sample was delivered to the laboratory. The vendor shall be responsible for holding times, preservation of the sample and the internal chain of custody from the time the vendor obtained the sample until the time the analysis is accepted by the Department. The vendor shall also maintain records of the results of analysis for a minimum of five (5) years.

### Step 2 -Conduct Specified Analysis on Samples

The methods used by the laboratory for the analysis shall be either; 1) Methods described in 40 CFR-136 and Standard Methods for the Examination of Water and Waste Water, current edition, but must be an approved method per 40 CFR Part 36 or. 2) Test Methods for Evaluating Solid Waste - Physical/Chemical Methods (SW -846) Third Edition with updates. The sampler shall be responsible for specifying either 1 or 2 above. In the event the method is not specified the laboratory shall contact the sampler for verification of the method to be used.

Analysis of samples is not deemed completed until the data has been submitted to and accepted by DWR and EE. Should the DWR and EE not provide notice of acceptance within four weeks of the date results were mailed, the firm may consider the data to be acceptable by the Division. The firm shall be responsible for maintaining preservation of the samples until the holding time is exceeded. Any samples with a sheen, discoloration or odor shall be maintained until DWR and EE's notification that the sample can be properly disposed of. DWR and EE will advise the firm which samples fall into this category. The firm shall be responsible for the proper disposal of all samples submitted to them by the DWR and EE unless otherwise notified. The firm shall dispose of the sample no earlier than four weeks after DWR and EE accepts the results. The results of the analysis shall be submitted to the DWR or EE no more than two (2) weeks after receipt of samples.

### Step 3 -Quality Control

Three programs are to be utilized to assure reliable laboratory data: (1) the use and documentation of standard analytical methods, (2) analysis of duplicate and spiked (where the method applies) samples at regular intervals each day to check analytical precision and accuracy, and (3) analysis of reference samples at 6 (six) month intervals\*. Regardless of which analytical methods are used in a laboratory, the methodology must be carefully documented. Standard methods that have been modified or entirely replaced because of recent advances in technologies may only be used when it has been given approval in the Federal Register. Documentation of procedures must be clear, honest, and adequately referenced; and the procedures shall be applied exactly as documented. The responsibility for results obtained from these procedures rests with the analyst and supervisor, both as representatives of the firm.

\*These analyses shall be conducted under the firm's performance evaluation test number through the Analytical Products Group.

To check the laboratory analytical precision, duplicate analysis of samples shall be performed at regular intervals. Duplicate samples must be carried through the complete analytical process. For all analyses, the interval shall be every tenth (10th) sample. When less than ten (10) samples are tested in one day, at least one duplicate sample shall be analyzed, and that sample must be a DWR or EE sample. The differences between the replicates for each analysis are to be plotted on Shewart precision quality control charts. "Out-of-Control" samples are to be repeated and appropriate steps shall be taken to locate and remedy the error.

To check the laboratory analytical accuracy, samples containing a known addition of the target analyte (spike) shall be analyzed at regular intervals. Spiked samples must be carried through the complete analytical process. For all analyses, the interval shall be every tenth (10th) sample. Where less than ten samples are tested in one day, at least one duplicate sample shall be analyzed, and that sample must be a DWR or EE sample. The percent recovery must be plotted out on Shewart accuracy quality control charts. "Out of Control" samples are to be repeated and appropriate steps taken to locate and remedy the source of error. The DWR and EE reserve the right to conduct unannounced examination of the laboratory's records to assure compliance.

Periodic submission of samples with known composition will occur. No notice of this activity will be provided unless results indicate an anomaly.

### Step 4. Legal Testimony

The selected firm or firms may be requested by the DWR or EE to testify concerning the validity of the laboratory analysis. The firm will only be required to testify to the following areas:

1. Time of notification by the Division of sampling and by whom.
2. When and where samples were collected by the firm.
3. Condition of sample.
4. How sample was preserved by the firm.
5. Date and time(s) of analysis and by whom.
6. Chain of Custody procedures within the laboratory.
7. Methods used.
8. Results of analysis.



At no time will the firm respond to questions concerning interpretation of results. The Division shall reimburse the firm for the costs of any such testimony.

## **Prime Vendor Responsibilities**

A vendor who is awarded a contract, when performing work under the terms and conditions of this contract, is solely responsible for the satisfactory completion of the work. The prime vendor shall be responsible for ensuring that any subcontractors have all the necessary permits, certifications (including WV State Laboratory Certification) and insurance to perform the work. DWR and EE will consider the prime vendor to be the sole point of contact with regard to authorized work under the contract, however this provision does not prohibit the DWR and EE from directly contacting subcontractors.

## **Subcontractors**

The prime vendor shall not be allowed to subcontract any work or services under this contract to any other person, company, corporation, firm, organization or agency without prior written approval of the DWR and EE.

## **Confidentiality**

The vendor agrees that any and all data, analyses, materials, reports or other information, oral or written, prepared by the vendor with respect to this requisition shall, except for information which has been publicly available, be treated as confidential and shall not be utilized, released, published, or disclosed, by the vendor at any time for any purpose whatsoever other than to provide consultation or other service to the DWR and EE.

## **Miscellaneous Provisions**

1. The DWR and EE will provide all sample containers and field preservatives.
2. The DWR and EE may, at their discretion, choose to deliver samples to the vendor's establishment rather than having them picked up by the vendor.
3. The DWR and EE reserve the right to deliver samples / portions of samples to firm(s) who are not awarded this analytical contract.
4. Upon awarding the contract, the vendor shall immediately provide at least 30 copies of the method detection limits (MDLS) for all analytes for which the contract is awarded. Any updates to the MDLS during the life of this contract shall be provided to the DWR and EE, in writing, within one week of the update(s) completion.
5. The firm shall provide at no additional cost, any requested quality control / calibration information associated with a particular sample. Quality control / calibration information includes but is not limited to: values of standards used in calibration, date of last calibration, correlation coefficients of calibrations curves, instrument blank values, check standard values, spike/recovery values, duplicate values, dilution volumes, bench sheets, calculations and Shewart quality control charts.

Category 1

nat. 1

Cost per test using:

Parameter	40 CFR 136 Methods for Liquid samples	40 CFR 136 Methods for Solid samples	SW-846 Methods for Liquid samples	SW-846 Methods for Solid samples
1 pH	3.00	5.00	3.00	5.00
2 Dissolved Oxygen	3.00	n/a	3.00	n/a
3 BOD	20.00	n/a	20.00	n/a
4 BOD-carbonaceous	20.00	n/a	20.00	n/a
5 Total Coliform (MF)	15.00	20.00	15.00	20.00
6 Total Coliform (MTF)	25.00	25.00	25.00	25.00
7 Fecal Coliform (MF)	15.00	20.00	15.00	20.00
8 Fecal Coliform (MTF)	25.00	25.00	25.00	25.00
9 Total Solids	10.00	10.00	10.00	10.00
10 Dissolved Solids	5.00	n/a	5.00	n/a
11 Suspended Solids	5.00	n/a	5.00	n/a
12 Volatile Solids	10.00	10.00	10.00	10.00
13 Percent Solids	5.00	5.00	5.00	5.00
14 Kjeldahl Nitrogen	20.00	20.00	20.00	20.00
15 Ammonia Nitrogen	20.00	20.00	20.00	20.00
16 Organic Nitrogen	20.00	20.00	20.00	20.00
17 Nitrate-Nitrogen	12.00	15.00	12.00	15.00
18 Nitrite-Nitrogen	12.00	15.00	12.00	15.00
19 Nitrite-Nitrate	12.00	15.00	12.00	15.00

## Category 2

Cat. 2

Cost per test using:

Parameter	40 CFR 136 Methods for Liquid samples	40 CFR 136 Methods for Solid samples	SW-846 Methods for Liquid samples	SW-846 Methods for Solid samples
1 Hot Acidity	5.00	n/a	5.00	n/a
2 Alkalinity	5.00	n/a	5.00	n/a
3 Hardness	14.00	24.00	14.00	24.00
4 Specific Conductance	2.50	5.00	2.50	5.00
5 Sulfate	8.00	10.00	8.00	10.00
6 Sulfide	10.00	n/a	10.00	n/a
7 Turbidity	5.00	n/a	5.00	n/a
8 Chloride	8.00	10.00	8.00	10.00
9 COD	20.00	n/a	20.00	n/a
10 TOC (subcontracted)	30.00	30.00	30.00	30.00
11 MBAS	20.00	n/a	20.00	n/a
12 Phenolics	25.00	25.00	25.00	25.00
13 Total Cyanide	25.00	25.00	25.00	25.00
14 Hexavalent Chromium	15.00	n/a	15.00	n/a
15 Oil-Grease	20.00	25.00	20.00	25.00
16 Fluoride	12.00	15.00	12.00	15.00
17 Total Phosphorus	10.00	10.00	10.00	10.00
18 Orthophosphate	8.00	8.00	8.00	8.00
19 Total Phosphate	8.00	8.00	8.00	8.00

Category 3  
Metals (Dissolved or Total)

Cost 3

High Level Detection

Flame Atomic Absorption / ICP

(dissolved metals will be field filtered)

Cost per test using:

Parameter      40 CFR 136 Methods for Liquid samples      40 CFR 136 Methods for Solid samples      SW-846 Methods for Liquid samples      SW-846 Methods for Solid samples

Parameter	40 CFR 136 Methods for Liquid samples	40 CFR 136 Methods for Solid samples	SW-846 Methods for Liquid samples	SW-846 Methods for Solid samples
1 Aluminum	5.00	13.00	5.00	13.00
2 Antimony	7.00	15.00	7.00	15.00
2 Arsenic	7.00	15.00	7.00	15.00
4 Barium	7.00	15.00	7.00	15.00
5 Beryllium	7.00	15.00	7.00	15.00
6 Boron (subcontracted)	15.00	20.00	15.00	20.00
7 Cadmium	7.00	15.00	7.00	15.00
8 Calcium	7.00	15.00	7.00	15.00
9 Chromium	7.00	15.00	7.00	15.00
10 Cobalt (subcontracted)	15.00	20.00	15.00	20.00
11 Copper	7.00	15.00	7.00	15.00
12 Iron	5.00	13.00	5.00	13.00
13 Lead	7.00	15.00	7.00	15.00
Magnesium	7.00	15.00	7.00	15.00
Manganese	5.00	13.00	5.00	13.00
16 Mercury (cold vapor)	20.00	20.00	20.00	20.00
Molybdenum	7.00	15.00	7.00	15.00
Nickel	7.00	15.00	7.00	15.00
19 Potassium	7.00	15.00	7.00	15.00
20 Selenium	7.00	15.00	7.00	15.00
Silver	7.00	15.00	7.00	15.00
Sodium	7.00	15.00	7.00	15.00
23 Thallium	7.00	15.00	7.00	15.00
Tin (subcontracted)	7.00	15.00	7.00	15.00
Vanadium	7.00	15.00	7.00	15.00
24 Zinc	7.00	15.00	7.00	15.00

Category 4

Metals (Dissolved or Total)

Low Level Detection

Graphite Furnace / ICP-MS

(dissolved metals will be field filtered)

*Cat 4*

Cost per test using:

Parameter                      40 CFR 136 Methods      40 CFR 136 Methods      SW-846 Methods      SW-846 Methods for  
   for Liquid samples      for Solid samples      for Liquid samples      Solid samples

1	Antimony	8.00	15.00	8.00	15.00
2	Arsenic	8.00	15.00	8.00	15.00
3	Beryllium	8.00	15.00	8.00	15.00
4	Boron	15.00	20.00	15.00	20.00
5	Cadmium	8.00	15.00	8.00	15.00
6	Chromium	8.00	15.00	8.00	15.00
7	Cobalt	15.00	20.00	15.00	20.00
8	Copper	8.00	15.00	8.00	15.00
9	Lead	8.00	15.00	8.00	15.00
10	Molybdenum	8.00	15.00	8.00	15.00
11	Nickel	8.00	15.00	8.00	15.00
12	Selenium	8.00	15.00	8.00	15.00
13	Silver	8.00	15.00	8.00	15.00
14	Thallium	8.00	15.00	8.00	15.00
15	Tin	8.00	15.00	8.00	15.00
16	Vanadium	8.00	15.00	8.00	15.00

Category 5

Miscellaneous Parameters

Cost / Analysis

Cost \$

1	Chlorophyll A (Standard Methods)	
2	Color (APHA)	
3	Color (ADMI)	
4	Cyanide, Amenable (40 CSR 136)	38.00
5	Cyanide, Free (ASTM)	38.00
6	Mineral Acidity (Standard Methods)	5.00
7	Total Acidity (Standard Methods)	5.00
8	Total Petroleum Hydrocarbons GRO/DRO (MW/TPH/8015B)	80.00
9	Fecal Streptococci (Standard Methods)	
10	Escherichia Coli (Numeric Result)	30.00
11	Bicarbonate (Standard Methods)	10.00
12	Ferrous Iron (Standard Methods)	15.00
13	Unionized Ammonia (46 CSR 1)	10.00
14	Dissolved Organic Carbon (Standard Methods) (subcontracted)	30.00
15	Particulate Organic Carbon (Standard Methods)	

Miscellaneous Expenses

Cost / hour

1 Professional staff representation of data in legal / administrative setting (hourly rate must include all travel expenses)

\$150.00

## Group 6 Constituents for Phase I Detection Monitoring<sup>1</sup>

### GROUP A: Inorganic Constituents

COMMON NAME <sup>2</sup>	CAS RN <sup>3</sup>	Bid as package
Acidity	(Total)	
Aluminum	(Total)	
Alkalinity	(Total)	
Ammonia Nitrogen	(Total)	
Antimony	(Total)	
Arsenic	(Total)	
Barium	(Total)	
Beryllium	(Total)	
Bicarbonates	(mg/l)	
Boron	(Total)	
Cadmium	(Total)	
Chlorides	(Total)	
Chromium	(Total)	
Cobalt	(Total)	
COD	(mg/l)	
Copper	(Total)	
Dissolved Manganese	(Total)	
Iron	(Total)	
Lead	(Total)	
Magnesium	(Total)	
Mercury	(Total)	
Molybdenum	(Total)	
Nickel	(Total)	
Nitrate	(Total)	
pH	(Std. Units)	
Potassium	(Total)	
Selenium	(Total)	
Silver	(Total)	
Sodium	(Total)	
Specific Conductance	(umhos/cm)	
Sulfate	(Total)	
TDS	(mg/l)	
Thallium	(Total)	
TOC	(mg/l)	
Total Phenolic Materials	(Total)	
TSS	(Total)	
Turbidity	(Total)	
Vanadium	(Total)	
Zinc	(Total)	

In addition to the above, the following parameters should be analyzed:

Temperature, (BOD-5-day), fluoride and calcium.

## Group 6 Constituents for Phase I Detection Monitoring<sup>1</sup> (continued)

### GROUP B: Organic Constituents

COMMON NAME <sup>2</sup>	CAS RN <sup>3</sup>
Acetone	67-64-1
Acrylonitrile	107-13-1
Benzene	71-43-2
Bromochloromethane	74-97-5
Bromodichloromethane	75-27-4
Bromoform; Tribromomethane	75-25-2
Carbon disulfide	75-15-0
Carbon tetrachloride	56-23-5
Chlorobenzene	108-90-7
Chloroethane; Ethyl chloride	75-00-3
Chloroform; Trichloromethane	67-66-3
Dibromochloromethane; Chlorodibromomethane	124-48-1
1,2-Dibromo-3-chloropropane; DBCP	96-12-8
1,2-Dibromoethane; Ethylene dibromide; EDB	106-93-4
o-Dichlorobenzene; 1,2-Dichlorobenzene	95-50-1
p-Dichlorobenzene; 1,4-Dichlorobenzene	106-46-7
trans-1,4-Dichloro-2-butene	110-57-6
1,1-Dichloroethane; Ethylidene chloride	75-34-3
1,2-Dichloroethane; Ethylene dichloride	107-06-2
1,1-Dichloroethylene; 1,1-Dichloroethene; Vinylidene chloride	75-35-4
cis-1,2-Dichloroethylene; cis-1,2-Dichloroethene	156-59-2
trans-1,2-Dichloroethylene; trans-1,2-Dichloroethene	156-60-5
1,2-Dichloropropane; Propylene dichloride	78-87-5
cis-1,3-Dichloropropene	10061-01-5
trans-1,3-Dichloropropene	10061-02-6
Ethylbenzene	100-41-4
2-Hexanone; Methyl butyl ketone	591-78-6
Methyl bromide; Bromomethane	74-83-9
Methyl chloride; Chloromethane	74-87-3
Methylene bromide; Dibromomethane	74-95-3
Methylene chloride; Dichloromethane	75-09-2
Methyl ethyl ketone; MEK; 2-Butanone	78-93-3
Methyl iodide; Iodomethane	74-88-4
4-Methyl-2-pentanone; Methyl isobutyl ketone	108-10-1
Styrene	100-42-5
1,1,1,2-Tetrachloroethane	630-20-6
1,1,2,2-Tetrachloroethane	79-34-5
Tetrachloroethylene; Perchloroethylene	127-18-4
Toluene	108-88-3
1,1,1-Trichloroethane; Methylchloroform	71-55-6



**Group 6 CONSTITUENTS FOR PHASE I DETECTION MONITORING<sup>1</sup> (continued)**

**GROUP B: Organic Constituents (continued)**

COMMON NAME <sup>2</sup>	CAS RN <sup>3</sup>
1,1,2-Trichloroethane	79-00-5
Trichloroethylene; Trichloroethene	79-01-6
Trichlorofluoromethane; CFC-II	75-69-4
1,2,3-Trichloropropane	96-18-4
Vinyl acetate	108-05-4
Vinyl chloride	75-01-4
Xylenes	1330-20-7

Cost for analysis of entire "Phase 1 Parameters" for landfills

\$ 700.00

1. This list contains volatile organics for which possible analytical procedures provided in EPA Report SW-846 "Test Methods for Evaluating Solid Waste", third edition, November 1986, as revised December 1987, includes Method 8260 and 8011; and metals for which SW-846 provides either Method 6010 or a method from the 7000 series of methods.

2. Common names are those widely used in government regulations, scientific publications, and commerce; synonyms exist for many chemicals.

3. Chemical Abstracts Service registry number. Where "Total" is entered, all species in the groundwater that contain this element are included.

## Collection of Samples

Cost associated with sample pickup from the following locations

		Cost / Collection
Charleston Offices	1356 Hansford St. Charleston, 24301	60.00
	1201 Greenbrier St. Charleston, 2301	60.00
Fairmont Office	2031 Pleasant Valley Rd. Fairmont 26554	150.00
Teays Office	PO Box 662 Teays, 25269	90.00
Romney Office	1 Depot St. Romney 26757	270.00
Wheeling Office	1060 Chapline St. Wheeling 26003	240.00
Parkersburg Office	2311 Ohio Ave. Parkersburg 26010	120.00
Oak Hill Office	116 Industrial Dr. Oak Hill 25901	30.00
Welch Office	311 Court St. Welch 24801	120.00
Logan Office	525 Tiller St. Logan 25601	150.00
Nitro Office	10 McJunkin Rd. Nitro 25143	90.00
Other Locations as necessary	Cost per mile to pickup site (example: pickup point is 50 miles from lab, if vendor bids \$X/mile, vendor charges \$50X, not \$100X)	1.30/mile



State of West Virginia  
 Department of Administration  
 Purchasing Division  
 2019 Washington Street East  
 Post Office Box 50130  
 Charleston, WV 25305-0130

# Purchase Order

**PURCHASE ORDER NO.:**  
 DEP12150A

**PAGE**  
 \_\_\_\_\_

**BLANKET RELEASE**  
 00

CORRECT PURCHASE ORDER NUMBER MUST APPEAR ON ALL PACKAGES, INVOICES, AND SHIPPING PAPERS. QUESTIONS CONCERNING THIS PURCHASE ORDER SHOULD BE DIRECTED TO THE BUYER AS NOTED BELOW.

**CHANGE ORDER**  
 \_\_\_\_\_

SEE REVERSE SIDE FOR TERMS AND CONDITIONS

**I N V O I C E T O**  
 ENVIRONMENTAL PROTECTION  
 DEPARTMENT OF  
 OFFICE OF ADMINISTRATION  
 10 MCJUNKIN ROAD  
 NITRO, WV  
 25143-2506

**V E N D O R**  
 \*331142322 304-255-4821  
 ANALABS INC  
 PO BOX 1235  
 CRAB ORCHARD WV 25827

**S H I P T O**  
 ENVIRONMENTAL PROTECTION  
 DEPARTMENT OF  
 OFFICE OF ADMINISTRATION  
 10 MCJUNKIN ROAD  
 NITRO, WV  
 25143-2506 304-759-0505

DATE PRINTED 01/30/2003	TERMS OF SALE NET 30	FEIN/SSN 550670153	FUND
SHIP VIA BEST WAY	F.O.B. DESTINATION	FREIGHT TERMS PREPAID	ACCOUNT NUMBER MUL-MUL

LINE	QUANTITY	UOP	VENDOR ITEM NO.	UNIT PRICE	AMOUNT
	DELIVERY DATE	CAT. NO.	ITEM NUMBER		
			RECEIPT TICKET FOR PURCHASE ORDER:		DEP12150A
LINE	CATNO	ITEM NUMBER	DESCRIPTION	QTY	DATE
0001		961-48	FIELD TESTING SERVICES		
		SIGNATURE _____		DATE _____	

IF APPROVAL AS TO FORM IS REQUIRED BY ATTORNEY GENERAL. CHECK HERE

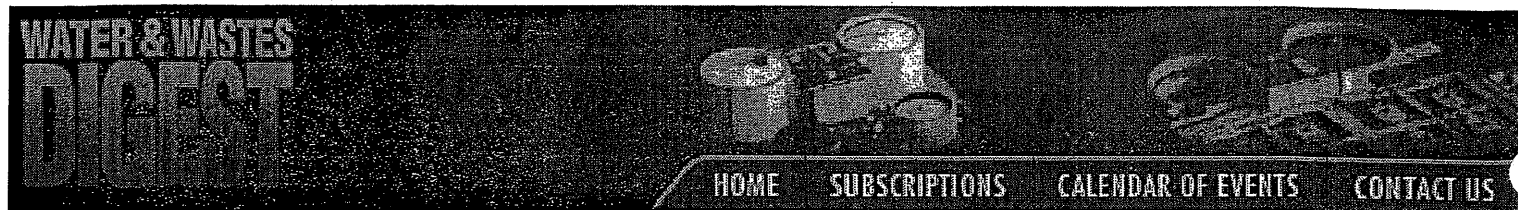
**TOTAL**

APPROVED AS TO FORM BY  
 ASSISTANT ATTORNEY GENERAL

BY \_\_\_\_\_  
 PURCHASING DIVISION AUTHORIZED SIGNATURE

**GENERAL TERMS & CONDITIONS  
PURCHASE ORDER/CONTRACT**

1. **ACCEPTANCE:** Seller shall be bound by this order and its terms and conditions upon receipt of this order.
2. **APPLICABLE LAW:** The laws of the State of West Virginia and the *Legislative Rules* of the Purchasing Division shall govern all rights and duties under the Contract, including without limitation the validity of this Purchase Order/Contract.
3. **NON-FUNDING:** All services performed or goods delivered under State Purchase Orders/Contracts are to be continued for the terms of the Purchase Order/Contract, contingent upon funds being appropriated by the Legislature or otherwise being made available. In the event funds are not appropriated or otherwise available for these services or goods, this Purchase Order/Contract becomes void and of no effect after June 30.
4. **COMPLIANCE:** Seller shall comply with all Federal, State and local laws, regulations and ordinances including, but not limited to, the prevailing wage rates of the WV Division of Labor.
5. **MODIFICATIONS:** This writing is the parties final expression of intent. No modification of this order shall be binding unless agreed to in writing by the Buyer.
6. **ASSIGNMENT:** Neither this Order nor any monies due, or to become due hereunder may be assigned by the Seller without the Buyer's consent.
7. **WARRANTY:** The Seller expressly warrants that the goods and/or services covered by this Order will: [a] conform to the specifications, drawings, samples or other description furnished or specified by the Buyer; [b] be merchantable and fit for the purpose intended; and/or [c] be free from defect in material and workmanship.
8. **CANCELLATION:** The Director of Purchasing may cancel any Purchase Order/Contract upon 30 days written notice to the Seller.
9. **SHIPPING, BILLING & PRICES:** Prices are those stated in this order. No price increase will be accepted without written authority from the Buyer. All goods or services shall be shipped on or before the date specified in this Order.
10. **LATE PAYMENTS:** Payments may only be made after the delivery of goods or services. Interest may be paid on late payments in accordance with the *West Virginia Code*.
11. **TAXES:** The State of West Virginia is exempt from Federal and State taxes and will not pay or reimburse such taxes.
12. **RENEWAL:** Any reference to automatic renewal is hereby deleted. The Contract may be renewed only upon mutual written agreement of the parties.
13. **BANKRUPTCY:** In the event the vendor / contractor files for bankruptcy protection, this contract is automatically null and void, and is terminated without further order.



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News this week sponsored by: YSI Inc

## Pittsburgh to Host Second National Water Quality Trading Conference

March 15, 2006

### INDUSTRY NEWS

The Second National Water Quality Trading Conference will be held May 23 to 25, 2006, in Pittsburgh, Pa. The event will highlight the economic and environmental benefits of water quality trading.


A collaboration of the U.S. Environmental Protection Agency and the U.S. Department of Agriculture and Farm Foundation, the intensive conference will focus on the policy and technical aspects of water quality trading. It will also explore opportunities and challenges associated with developing trading programs.

Source: EPA March 15, 2006

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- [Former USDA Plant Pathologist Randy J. McLaughlin, Ph.D. Joins Novazone](#)
- [PADEP Awards Statewide Environmental Contract to alfred benesch & company](#)

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- [Global Residential Water Treatment Equipment Market Creates \\$15 Billion Opportunity](#)
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Appendix C-2

Contract Specifications

For

Macroinvertebrate Sample

Processing and Identification

May 27, 2004  
**CONTRACT SPECIFICATIONS FOR  
BENTHIC MACROINVERTEBRATE SAMPLE PROCESSING  
AND/OR IDENTIFICATION**

**AREA OF WORK/BID AWARD**

The Department of Environmental Protection, Division of Water and Waste Management is seeking bids for the processing and identification of benthic macroinvertebrates collected from West Virginia waters. Macroinvertebrates will be collected from riffle / run habitats of flowing waters using rectangular dip nets.

Bids should be submitted by vendors in connection with the following:

- Sample pick-up and delivery
- Removal of organisms from stream debris
- Identification of the sample to the genus level and electronic submission of results
- Legal representation (\$/hour)
- Quality assurance / quality control for sample processing and identification

Bid awards may be made to more than one vendor. The program typically collects around 550 samples a year, but no minimum volume of samples is guaranteed to any one vendor.

Additional information that is mandatory for the bid submittal is found in the Proposal Preparation section of this solicitation.

**QUALIFICATIONS**

The Department of Environmental Protection's (DEP) Division of Water and Waste Management (DWWM) conducts inspections of permitted and non-permitted facilities, investigates complaints, monitors ambient quality of surface water, groundwater and sediments, performs studies, and provides water quality information to the citizens of West Virginia and other government agencies.

DWWM's Watershed Assessment Section (WAS) performs the majority of the macroinvertebrate sampling. WAS has collected an average of 550 benthic macroinvertebrate samples annually; most samples are collected between April and October each year.

Legal action based upon identification results is possible. Therefore, the firm or firms selected shall have a quality control program in place and shall meet the following qualifications:



1. Must have degreed biologists on staff **that perform the actual identifications.**  
(Identification of organisms by non-professional personnel is strictly forbidden.)  
Biologists must be dedicated taxonomists; that is, the majority of the work performed involves the identification of aquatic organisms.
2. Must be capable of attending and providing expert testimony in legal proceedings, upon request.
3. Must submit results for completed samples bimonthly (every two months). Bimonthly submissions shall include the following for each sample: a) macroinvertebrate identifications in Excel or Access format; b) copies of bench sheets; c) all organisms identified (except reference specimens, which will be returned when the contract is complete); and d) all QA/QC associated with sorting and identifying the samples.
4. Must be able to complete entire project by Feb. 28<sup>th</sup> of the year following the dates of collection (for all samples collected prior to October 15<sup>th</sup>). Results of smaller, site-specific projects must be available within one month of sample receipt or within some other negotiated time period.
5. Must be able to demonstrate success in completing large-scale macroinvertebrate processing and identification projects (e.g., projects with >200 samples per year).

## **SCOPE**

In administering and enforcing most of the pollution control laws of the state, the importance of quality control cannot be overstated. Quality control measures must be strictly adhered to in all phases of sample collection, preservation, transportation, and analysis. The quality control and analytical processes, as they relate to the contractor's responsibility, are divided into four (4) major steps:

STEP 1 - Collection of sample from specified office.

STEP 2 - Conduct specified analysis on samples in a timely and professional manner.

STEP 3 - Establishment of continuing program to ensure the reliability of data (Quality Assurance/Quality Control).

STEP 4 - Legal Testimony

### Step 1 - Collection of Samples from Specified Office

Benthic macroinvertebrate samples will be collected by DWWM personnel. Due the size of the

sample containers (1 gallon Nalgene) and the total number of samples collected annually, DWWM will not ship samples to the contractor using commercial transport such as UPS or Federal Express. Therefore, the vendor shall provide sample pick up and delivery services. DWWM will bear the cost of sample transport, however, and the vendor shall include sample transport costs as part of the bid package. Typically, there are four to five sample pick-ups per year.

DWWM will provide Chain-of-Custody forms when samples are picked-up by the vendor. The vendor shall be responsible for maintaining preservation of the sample and the internal chain of custody from the time the vendor obtains the sample until the results of macroinvertebrate identification are accepted by the Division. The vendor shall also maintain records of the results of identification for a minimum of five (5) years.

## Step 2 - Conduct Specified Analysis on Samples

### Sorting Benthic Macroinvertebrate Samples

Benthic macroinvertebrate samples shall be processed in accordance with the procedures outlined in "Standard Operating Procedures for Processing Benthic Macroinvertebrate Samples" (Attachment A.). Sub-samples consisting of 200 aquatic macroinvertebrates are to be prepared for all samples collected with a net apparatus. Sub-samples shall be obtained by placing the entire sample in a sieve box divided into 100 1-inch by 1-inch grids. Any vertebrates encountered during subsampling should be retained with the sample, but not identified. Specimens should be stored in archival quality containers that will prevent loss of preservative through evaporation: glass vials with or without screw caps, polypropylene jars with screw caps, etc.

Vendor will be responsible for examining sorting efficiency for 5 % of all submitted samples for QA/QC purposes.

### Identification of Benthic Macroinvertebrate Samples

Benthic macroinvertebrate samples shall be identified in accordance with procedures outlined in "Standard Operating Procedures for Identifying Macroinvertebrate Samples" (Attachment B.). Taxonomists are permitted to use identification keys other than those suggested in the operating procedures. However, all keys must be current and up-to-date. All results submitted to DWWM shall include a bibliography of publications used in identification of the specimens. Vendor will be responsible for identification only; data analysis will not be required.

All aquatic insects (including Diptera), crayfish, snails and clams are to be identified to the genus-level. ***(NOTE: samples may include a significant number of chironomid larvae, which MUST be identified to genus.)*** Aquatic invertebrates that do not require family/genus level identification are Nemertea, Oligochaeta, Nematoda, Hydroida, Turbellaria, Bryozoa, and Hirudinea. These organisms need only be identified to the taxonomic level (phylum, class, order, etc.) indicated in the previous sentence. Vertebrates and terrestrial organisms are not to be identified.

Vendor will be responsible for re-identification of 5 % of all submitted samples for QA/QC purposes.

Vendor will also be required to establish reference collections and retain all voucher specimens for this project. A reference collection is defined as a set of biological specimens, each representing some taxonomic level. A reference collection is not necessarily limited to a single watershed assessment. Reference collections are to be arranged/curated based on taxonomic and/or phylogenetic order. Voucher specimens are the actual specimens collected during a watershed assessment.

Results of identifications shall be submitted on the form(s) provided by DWWM and in electronic format (Microsoft Excel or Access compatible format; WVDEP will provide a blank database for this purpose). Entire project must be completed by February 28 of the year subsequent to that of the sample collection. However, vendor must submit data for completed samples bimonthly (every two months). Bimonthly submissions will include the following: a) macroinvertebrate identifications in electronic format; b) bench sheets; c) all macroinvertebrate specimens (except reference materials); d) all QA/QC associated with sorting and identification of each sample. DWWM and the vendor may negotiate an earlier completion date upon the mutual agreement of both parties.

Analysis of samples is not deemed completed until the data has been submitted to and accepted by the DWWM. Should the DWWM not provide notice of acceptance within four weeks of the date results were mailed by the vendor, the firm may consider the data to be acceptable by the Division.

The vendor shall be responsible for maintaining preservation of the samples. Vendor shall return all sample jars, voucher specimens and reference collections to the DWWM in addition to the results of identification. Unused sample residues (i.e. detritus and unpicked portions) are to be properly disposed by the vendor.

### Step 3 - Quality Control

The consultant shall compile genus-level reference and voucher collections to be submitted to DEP/DWWM at the completion of the project.

With the exception of organisms used in the reference collection, all specimens identified in the 200 organism sub-samples are to be returned to DEP/DWWM. Slide mounted specimens should be labeled to indicate, at a minimum, DEP sample ID and lab number. All other specimens are to be stored in a single sample vial (additional vials may be used when large organisms, (i.e., crayfish) are present).

Vendor shall evaluate sorting efficiency for 5 % of all samples. Recovery errors may not exceed 10% of the total sample. A record of all samples sorted, a list of quality control checks and documentation of any corrective action taken shall be maintained by the vendor to document the process. This information shall be provided bimonthly.

In addition, the vendor shall re-identify a minimum of 5 % of the samples. A taxonomist other than the original identifier shall perform this check. All documentation associated with the QA/QC process, including any corrective action taken, shall be submitted to DEP/ DWWM bimonthly.

DEP biologists and/or another contract laboratory will verify identifications for a minimum of 2.5% of the samples. Samples subjected to verification are selected randomly and will encompass checks on all taxonomists. The vendor will be advised immediately if significant<sup>1</sup> differences in identification are encountered. Cancellation of the contract will result if discrepancies continue.

### Step 4 - Legal Testimony

The selected firm or firms may be requested by the DWWM to testify concerning the validity of the laboratory analysis. The firm will only be required to testify to the following areas:

1. Time of notification by the DEP/DWWM of sample shipment and by whom.
2. Condition of sample.
3. How sample was preserved by the firm.
4. Dates of analysis and by whom.

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<sup>1</sup> "Significant" differences will include, but will not be limited to, consistent misidentification of an organism(s) during QA/QC checks.

5. Chain of Custody procedures within the laboratory.
6. Methods used.
7. Results of analysis.

At no time will the firm respond to questions concerning interpretation of results. The Division shall reimburse the firm for the costs of any such testimony.

### **PRIME VENDOR RESPONSIBILITIES**

The vendors who are awarded a contract, when performing work under the terms and conditions of this contract, are solely responsible for the satisfactory completion of the work. The prime vendor shall be responsible for ensuring that any subcontractor has all the necessary permits, certifications, experience and insurance to perform the work. All subcontractors must be pre-approved by DWWM before subcontractor initiates work. The primary contractor shall supply resumes and/or other documents to prove sub-contractor's qualifications. DWWM will consider the prime vendor to be the sole point of contact with regard to authorized work under the contract; however, this provision does not prohibit the DWWM from directly contacting subcontractors.

### **CONFIDENTIALITY**

The vendor agrees that any and all data, analyses, materials, reports or other information, oral or written, prepared by the vendor with respect to this requisition shall, except for information which has been publicly available, be treated as confidential and shall not be utilized, released, published, or disclosed, by the vendor at any time for any purpose whatsoever other than to provide consultation or other service to the DWWM.

## PROPOSAL PREPARATION

In the proposal the bidder shall include the information listed below. DWWM will evaluate this information prior to recommending a contract award, which will be made to the three qualified vendors with the lowest bids as determined from the 'Bid Costs Evaluation Formula'. We expect to collect approximately 550 samples, however there is no minimum number of samples that will be sent to any one of the successful bidders. **Omission of any of the information listed below may result in disqualification.** Three vendors are necessary to ensure that DWWM has alternate vendors to perform additional QA/QC reviews.

- 1) Description of how the project will be managed by the contractor.
- 2) Summary of experience with sorting and identification of benthic macroinvertebrates. Resumes of taxonomists shall be included in the bid package.
- 3) Specific description of how the reference collection will be made.
- 4) Specific description of sorting procedures, including sub-sampling protocols.
- 5) Specific description of methods used to prepare Chironomidae for identification.
- 6) List of taxonomic references used in the identification of all specimens.
- 7) Description of vendor's internal QA/QC procedures, stating specifically how errors are resolved, which will insure the highest level of accuracy in both the sorting and identifying processes.
- 8) Specific description of product that will be returned to DEP/DWWM (ie, reporting format, specimens, etc.)
- 9) Bids must include :
  - A.\$ \_\_\_\_\_ Per sample un-sorted, identified to genus level
  - B \$ \_\_\_\_\_ Per sample pre-sorted, identified to genus level (necessary for quality assurance checks)
  - C \$ \_\_\_\_\_ Per each sample pick-up/delivery (assume 100 samples per pickup)
  - D \$ \_\_\_\_\_ Cost/hr for professional staff representation of data in legal/administrative setting

Contact Person: \_\_\_\_\_

Contact Phone #: \_\_\_\_\_

\* Bid Costs Evaluation Formula':  $Bid = A + B + (C/30) + (D/20)$   
 (un-sorted genus level ID) + (sorted genus level ID) + (sample pickup / delivery/30) + (legal rep/20)

**List of Prospective Vendors:**

There are few individuals/companies that perform the services described in the contract specifications. A few potential vendors are listed below.

James B. (Sam) Stribling, Ph. D.  
Tetra Tech, Inc.  
10045 Red Run Blvd., Suite 110  
Owings Mills, MD 21117-6102  
Phone (410) 356-8993  
Fax (410) 356-9005  
E-mail: [jamstr@ccpl.carr.lib.md.us](mailto:jamstr@ccpl.carr.lib.md.us)

Mike Winnell  
Freshwater Benthic Services  
3250 Krause Road  
Petoskey, MI 49770  
Phone and Fax: 231-347-9752.  
E-mail: [mwinnell@freeway.net](mailto:mwinnell@freeway.net)

Wendell Pennington  
Pennington & Associates, Inc.  
Suite 103  
Cookeville, TN 38501  
Phone: (913) 526-6038

Molly Foree  
Third Rock Consultants, LLC.  
2514 Regency Road  
Suite 104  
Lexington, KY 40503  
Phone: (859) 977-2000

Gary T. Lester  
EcoAnalysts, Inc.  
105 East 2nd Street, Suite 1  
Moscow, ID 83843  
Phone (208) 882-2588, fax (208) 883-4288

Stuart Linde  
Environmental Services & Consulting  
101 Professional Park Dr. Suite 303  
Blacksburg, VA 24060  
(540) 552-0144

John Johanson  
Appalachian Technical Services  
6741 Indian Creek Rd  
Wise, Virginia 24293  
(276)328-4200

Consolidated Environmental Engineering  
245 East Dr.  
570 E. 10<sup>th</sup> Street  
Melbourne, FL 32904

GAI Consultants  
3412 Chesterfield Ave  
Charleston WV 25143

Environmental Solutions & Innovations  
781 Neeb Rd.  
Cincinnati, OH 45233  
(513) 451-1777

REIC Laboratory  
PO Box 286  
Beaver WV 25813

**Attachment A.**

**WVDEP/DWWM Requirements for Processing Benthic Macroinvertebrate Samples (Preparing a 200 organism sub-sample)**

**INTRODUCTION**

Sorting macroinvertebrates from benthic survey samples (a procedure often referred to as "bug sorting") is an extremely important step in the biological research performed by the Department of Environmental Protection. The quality of the work performed by the "sorter" influences the quality of subsequent processes, such as identification and data analysis. A competent "sorter" must be able to recognize the morphological diversity of aquatic organisms, as well as the various methods these organisms may use to hide themselves from predators. The outcome of the final study may be affected, even if only a few organisms are overlooked during the sorting process.

The processes described below were derived from: Barbour, M. T. et al. "Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish", Second Ed., EPA 841-B-99-002. These protocols may be downloaded from the Internet at <http://www.epa.gov/owowwtr1/monitoring/rbp/download.html>.

**DEFINITIONS**

**MACROINVERTEBRATES** - Animals that are large enough to be seen with the naked eye and do not have a backbone.

**BENTHIC ORGANISMS (or BENTHOS)** - Living organisms that reside on the bottom of streams, rivers, or lakes. Benthos may be vertebrates, invertebrates, or plants.

**REFERENCE COLLECTION** - A reference collection is a set of specimens, each representing some taxonomic level and not necessarily limited to a specific project. For the purposes of DWWM's studies, a reference collection does not have to be limited to a particular watershed. Reference collections should have expert confirmation of each taxon. These collections are used to verify identifications of subsequent samples.

**VOUCHER COLLECTION** - The voucher collection consists of the actual specimens collected during the project. Following identification and enumeration, all specimens collected for this project should be maintained in a voucher collection. This collection will be returned to the DWWM.



## EQUIPMENT

1. Sample Jar - Contains the unprocessed sample, which consists of benthic organisms and stream debris.
2. Sample Bottle - for storage of processed sample. This container may be a plastic/glass vial or a larger plastic bottle. Most samples will fit into a 10 ml vial, however, large organisms such as crayfish, will require larger bottles.
3. White Flat-bottom Pans - contain sample during the sorting process
4. Denatured Alcohol - preservative used in unprocessed and processed samples
5. Sieves - #30 sieves are used to separate alcohol and fine debris from the sample prior to sorting.
6. Sieve box - a homemade wooden frame with #30 mesh screening on the bottom is used to evenly distribute the sieved sample for randomly selecting the sub-sample. The internal dimensions of the box can vary (i.e., 10 in. X 10 in. or 5 in. X 20 in.); however, all boxes are marked into 100 1-inch by 1-inch grids.
7. Labels - Self-adhesive labels are used to identify the contents of the sample bottle (i.e., the sorted sample).
8. Scotch Tape - Used on label as additional adhesive.
9. Pencil - used to label sample bottle.
10. Crucible - or other small container, is used for short term, intermediate storage of the sample during the sorting process.
11. Forceps - Fine tipped forceps are used to remove the organisms from the debris.
12. Illuminated Magnifier - an optical aid to illuminate and magnify the sample during the sorting process. Alternatively, magnifying visors and a desk lamp can be used.
13. Squirt bottle - filled with alcohol, used to rinse organisms into sample bottle.
14. Plexiglas - used to cover sample partially sorted overnight to prevent evaporation.

## SAFETY

Protective eyewear should be worn during sample processing to prevent contact with the residual alcohol in the specimens and debris.

## PROCEDURES

1. Select the sample to be sorted. If a sample is in two jars, the contents of the jars must be combined before picking is initiated.
2. Select a small bottle that will hold the organisms after sorting is completed. Usually a 10 mL bottle is adequate for a 200-organisms sub-sample. An additional bottle may be needed if the sample contains large organisms.
3. Label the bottle:
  - a. Use self-adhesive labels
  - b. Using a pencil (ink will run if alcohol is spilled on the label), copy all information on the sample jar label onto the self-adhesive label. The label must include the following information:

Stream Name  
Station Number (Random Number and/or AN-code)  
Sample ID# (Short number to link electronic results to final database)  
Sample Date  
County  
Initials of Sample Collector  
Initials of Sample Processor  
# of grids sorted  
# of organisms in final sample

If any of this information is missing from the original sample jar label, notify the DWWM biologists so that the error can be corrected.

- c. Stick the new label on the bottle and secure with clear tape.
  - d. Prepare an *internal* label for the sample using permanent ink. Internal label should contain the same information as required in 3-b above. This label will serve as a back-up if the external label is lost. (External labels may be omitted for samples stored in

transparent glass containers, as long as the internal label is clearly visible.)

4. Prepare the sample for sorting. This step is performed in a sink and should be done under a fume hood or in a well ventilated area.

- a. Under a fume hood, open sample jar and pour contents into the sieve box
- b. Rinse sample jar into sieve box and examine jar to make sure all detritus and bugs have been removed.
- c. Rinse the contents of the sieve box in tap water to remove remaining alcohol and to rinse out fine sand and sediment.
- d. Carefully rinse any large detritus (i.e. leaves) or stones, making sure that all organisms on these items are returned to the sieve. Discard the leaves and rocks after rinsing.
- e. Place the box in a few inches of water and gently swirl it until the contents are evenly distributed. ***Even distribution is extremely important in this step.*** If debris is clumped, the organisms will not be distributed evenly and the final result may be skewed. If the sample was divided into more than one jar, the jars are to be combined at this point. When the sample is evenly distributed throughout the gridded screen box, remove it from the water.
- f. Using a random number generator, select the first grid to be sorted. Using the "cookie cutter", isolate the organisms within the chosen grid and scoop the contents of the grid into a white enamel pan. Be careful not to destroy any organisms during this step. Organisms with their head inside the grid are to be included within the grid. If you can't tell which end is the head, then the organism belongs in the grid that contains the largest portion of the body.

#### 5. Sorting

- a. Fill a crucible with 75% alcohol. A small piece of tape, rolled into a ring so the adhesive is exposed, may be attached to the bottom of the crucible to prevent tipping. If preferred, another small wide-mouth container may be substituted for the crucible.
- b. Using fine-tipped forceps and illuminated magnifier or magni-visor (sorter should use magnification of at least 2x), remove all invertebrates from the sub-sample and transfer to the alcohol filled crucible. Keep track of the number of organisms that have been sorted. ***If there are a significant number of invertebrates that appear to be terrestrial, include them in the sample, but do not include them in the 200-organism count. The taxonomist will verify whether these organisms are truly terrestrial or semi-aquatic. Do***

*not include empty clam or snail shells, or parts of organisms that are easily disconnected from the specimen (legs, gills, ect.).*

- c. If leaves are present, be sure to examine both surfaces. Watch for unusual clumps of twigs, leaves, or sand, which may be protective cases for some organisms. If cases are found, both the case and the organism should be sorted. If the organism is in the case, the case and organism should be kept together. If an empty case is found, it should also be removed.
- d. If there is any doubt to the identity of an object (is it a seed or a bug?), it should be sorted, but not counted. A senior biologist should be notified if a large number of questionable objects are present.
- e. When all the organisms appear to have been removed from the pan, agitate the contents of the pan and look again. Often the agitation will reorient an organism that was previously overlooked.
- f. Have a senior biologist inspect the pan after sorting has been completed. The biologist will point out any organisms that have been overlooked or misidentified as detritus. As the sorter becomes more proficient at his/her task, this step will be reduced in frequency.
- g. If 200 or more organisms have been obtained from the initial grid chosen, sub-sampling is complete. If fewer than 180 organisms have been collected, another grid is randomly chosen and steps 4.f through 5.e are repeated until at least 180 organisms are obtained or until the entire sample has been sorted. The remainder of the sample (i.e., the non-selected grids) may be discarded.
- h. Pour the contents of the crucible into the labeled bottle. Use a squirt bottle containing alcohol to rinse the organisms from the crucible. Make sure that all organisms in the bottle are fully submerged in the alcohol and that none are clinging to the sides of the bottle. Use the squirt bottle to rinse the sides of the bottle, necessary.

## 6. Record Keeping

- a. After a sample has been sorted, record the date and your initials in the sample log book. **The total number of organisms picked and the number of grids sorted** should also be documented for each sample. This last step is very important as these values are used to calculate organism density and to determine sample comparability.

## **QUALITY ASSURANCE/QUALITY CONTROL**

Sorting efficiency shall be evaluated for 5% of the samples. Recovery errors cannot exceed 10% (i.e., no more than 20 organisms can be missed by the sorter for a given sample) of the total sample (composite of remnants from each grid sorted). If the sorter does not meet this standard, the sorted sample remnants shall be re-checked until the recovery limits are attained. A record of all samples sorted, a list of quality control (QC) checks and documentation of any corrective action taken shall be maintained by the vendor to document the process. DWWM reserves the right to review QA/QC documentation upon request.

## Attachment B.

### WVDEP/DWWM Requirements for the Identification of Macroinvertebrates

Consultant will be required to provide identification services only. No data analysis will be required. At the completion of the projects (or portions of the project) the consultant will submit the completed "WVDEP/ WAS BENTHIC MACROINVERTEBRATE LAB SHEET", voucher specimens, and identification results in Excel or Access format. (The voucher specimens are essentially all specimens in the 200 organism sub-sample that have not been included in the reference collection.) Vendor may retain reference specimens until the project has been completed.

Aquatic insects (*including all Diptera*), crayfish, snails and clams in the 200-organism subsample are to be identified to the genus level as specified in the project contract. Aquatic invertebrates that do not require genus level identification are Nemertea, Oligochaeta, Nematoda, Hydroida, Turbellaria, Bryozoa, and Hirudinea. These organisms need only be identified to the taxonomic level (phylum, class, order, etc.) indicated in the previous sentence. Vertebrates and terrestrial organisms are not to be identified. If these organisms are included in the sample, they shall be retained with the sample and returned to DWWM.

### MATERIALS AND SUPPLIES

1. Dissecting Microscope - for examination of gross features.
2. Compound Microscope - for examining minute features. Phase-contrast microscopes are preferable.
3. Fine-tipped forceps - for manipulating specimens.
4. Fine-tipped probes - for manipulating specimens.
5. Petri dishes – or other container to hold specimens during identification.
6. Alcohol - 75% ethanol or isopropanol is used to preserve the samples and to prevent desiccation during identification.
7. Wash Bottle - used for alcohol storage.
8. Microscope Slides and glass cover slips - for examination of tiny specimens and/or body parts under a compound microscope. Slides and cover slips should be clean.

9. Benthic Macroinvertebrate Lab Sheet - standard for recording results of identification and enumeration (Figure 1).
10. Mounting Medium – CMC-10 mounting medium is used to prepare permanent mounts of microscopic specimens.





## 11. Taxonomic Keys –

The primary taxonomic keys are listed below. The contractor may use other taxonomic keys for lower level identification; however, these references must be current and up-to-date. The contractor shall provide a list of references used in the identification of all specimens.

Merritt, R. W. and K. W. Cummins, eds. 1996. An Introduction to the Aquatic Insects of North America, Third Edition. Kendall and Hunt Publishing Company, Debuque, Iowa.

Pennack, R. W. 1978. Fresh-water invertebrates of the United States. John Wiley and Sons, New York

Brown, H. P. 1972. Aquatic dryopoid beetles (Coleoptera) of the United States. U. S. Government Printing Office.

Burch, J. B. 1982. Freshwater Snails (Mollusca: Gastropoda) of North America. United States Environmental Protection Agency. EPA-600/3-82-026.

Edmunds, G. F., Jr., S. L. Jensen, and L. Berner. 1976. Mayflies of North and Central America. University of Minnesota Press.

McAlpine, J.F. (Ed.). 1989. Manual of Nearctic Diptera. Vols. 1-3. Research Branch Agriculture Canada. Monograph No. 32.

Needham, J. G. et al. 2000. Dragonflies of North America. Scientific Publishers.

Peckarsky, B. L., P. R. Fraissinet, M. A. Penton, and D. J. Conklin, Jr. 1990. Freshwater Macroinvertebrates of Northeastern North America. Cornell University Press.

Stewart, K. W. and B. P. Stark. 1988. Nymphs of North American Stonefly Genera (Plecoptera). Entomological Society of America.

Thorp, J.H and A.P.Covich, Eds. 2001. Ecology and Classification of North American Freshwater Invertebrates. Second Edition. Academic Press.

Westfall, M.J. and M. L. May. 1996. Damselflies of North America. Scientific Publishers.

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and South Carolina. Available on-line:

[http://www.esb.enr.state.nc.us/BAUwww/Chiron\\_manual/intro.pdf](http://www.esb.enr.state.nc.us/BAUwww/Chiron_manual/intro.pdf)

### **Procedures for mounting Chironomidae ( and other small specimens)**

The procedures that follow are summarized from Epler's *Identification Manual for the Larval Chironomidae (Diptera) of North and South Carolina*.

1. Label a clean glass slide. Label should include, at a minimum, the stream name, stream code, collection data and sample ID number.
2. Place 2-5 drops of CMC-10 mounting medium on the slide.
3. Place the specimens in the mounting medium, ventral side up, head pointing down ("south"). Tease out larger bubbles.
4. Gently lower coverslip over the mounting medium at an angle.
5. Use the cover slip to reposition larvae, if desired. Then gently press down the cover slip over the head capsules with pencil eraser to spread the mouthparts and over the anal end to spread the hind pro-legs.
6. Lay the slide on a flat surface and allow it to cure for 2-3 hours. If air bubble form, fill them in with fresh medium and allow to cure 1-2 more hours. Then ring the slide with more medium or clear fingernail polish.

### **Quality Assurance / Quality Control**

A minimum of 5 percent of the samples are re-identified by a taxonomist other than the original identifier. Errors are brought to the attention of the original taxonomist and subsequent identifications are subject to scrutiny until errors are resolved.

Appendix C-3

Contract Specifications

For

Periphyton Identification

**#DEP13585A**  
**CONTRACT SPECIFICATIONS FOR PERIPHYTON SAMPLE  
PROCESSING AND IDENTIFICATION**

**AREA OF WORK / BID AWARD**

The West Virginia Department of Environmental Protection, Division of Water and Waste Management (DWWM) is seeking bids for the processing and identification of periphyton samples collected from streams of West Virginia. Personnel from DWWM's Watershed Assessment Section will collect and preserve the samples. There are typically between 250 and 350 samples collected each year that would need processed and identified. As of 12/2005, we have a backlog of around 200 samples, which would be shipped upon as soon as contract is signed.

Bids should be submitted by vendors in connection with the costs associated with processing (including cleaning and preparation of slides for diatoms) and identification of the periphyton samples.

Additional information that is mandatory for the bid submittal is found in the Proposal Preparation section of this solicitation.

**QUALIFICATIONS**

The Department of Environmental Protection's (DEP) Division of Water and Waste Management (DWWM) conducts inspections of permitted and non-permitted facilities, investigates complaints, monitors ambient quality of surface water, groundwater and sediments, performs studies, and provides water quality information to the citizens of West Virginia and other government agencies. Legal action based upon identification results is possible. Therefore, the firm or firms selected must have a quality control program in place and meet the following qualifications:

1. Degreed biologist on staff **who performs the actual identifications.**  
(Identification of organisms by non-professional personnel is strictly forbidden)
2. Capable of attending and providing expert testimony in legal proceedings, upon request.
3. Capable of processing and identifying 30 samples per month.

**SCOPE**

In administering and enforcing most of the pollution control laws of the state, the importance of quality control cannot be overstated. Quality control measures must be strictly adhered to in all phases of sample collection, preservation, transportation, and analysis. The quality control and analytical processes, as they relate to the contractor's responsibility, are divided into four (4) major steps:

STEP 1 - Collection of sample from specified office.

STEP 2 - Conduct specified analysis on samples in a timely and professional  
~~STEP 3~~ - Establishment of continuing program to ensure the reliability of data  
(Quality Assurance/Quality Control).  
STEP 4 - Legal Testimony

### **Step 1 - Collection of Samples from Specified Office**

Collection of periphyton samples shall be conducted by DWWM personnel. Each sample will be a 100 ml graduated container (sealed w/ electrical tape) with periphyton scraped from 5 rocks mixed with rinse water and preserved with formalin. These will generally be total samples. There will be some split for QA purposes. The vendor will be notified of sample shipment. Costs of sample shipment to the vendor will be borne by the DWWM. Costs to return identified slides and results to the DWWM will be the sole responsibility of the successful bidder(s). The vendor shall be responsible for preservation of the sample and the internal chain of custody from the time the vendor obtains the sample until the time the analysis is accepted by the Division. The vendor shall also maintain records of the results of identification for a minimum of three (3) years.

### **Step 2 - Conduct Specified Analysis on Samples**

Processing and Identification of Periphyton Samples shall be carried out according to vendor's procedures as defined in response to this request

Results of identifications shall be submitted to DWWM at a rate of at least 30 samples per month, starting 30 days from the receipt of samples or at an alternate rate that is determined acceptable by DWWM.

Analysis of samples is not deemed completed until the data has been submitted to and accepted by the DWWM. Should the DWWM not provide notice of acceptance within four weeks of the date results were mailed by the vendor, the firm may consider the data to be acceptable by the Division.

### **Step 3 - Quality Control**

Quality control procedures should be well defined and strictly adhered to in all aspects of processing, storage, and identification. Quality control procedures must be submitted as part of this bidding process. Any cost for internal QA/QC procedures should be incorporated into the cost / sample bid.

### **Step 4 - Legal Testimony**

The selected firm or firms may be requested by the DWWM to testify concerning the validity of the laboratory analysis. The firm will only be required to testify to the following areas:

1. Time of notification by the Division of sample shipment and by whom.
2. Condition of sample.
3. Date and time(s) of analysis and by whom.
4. Chain of Custody procedures within the laboratory.
5. Methods used.
6. Results of analysis.

The Division shall reimburse the firm for the costs of any such testimony.

## **PRIME VENDOR RESPONSIBILITIES**

A vendor, who is awarded a contract, when performing work under the terms and conditions of this contract, is solely responsible for the satisfactory completion of the work. The prime vendor shall be responsible for ensuring that any subcontractors have all the necessary permits, certifications, experience and insurance to perform the work. DWWM will consider the prime vendor to be the sole point of contact with regard to authorized work under the contract; however, this provision does not prohibit the DWWM from directly contacting subcontractors.

## **CONFIDENTIALITY**

The vendor agrees that any and all data, analyses, materials, reports or other information, oral or written, prepared by the vendor with respect to this requisition shall, except for information which has been publicly available, be treated as confidential and shall not be utilized, released, published, or disclosed, by the vendor at any time for any purpose whatsoever other than to provide consultation or other service to the DWWM.

## **PROPOSAL PREPARATION**

The bidder shall include the following information. DWWM will evaluate this information prior to recommending a contract award.

- 1) Description of how the project will be managed by the contractor.
- 2) Summary of experience with sorting and identification of periphyton.
- 3) Description of how both "soft" algae and diatoms will be processed and identified, while meeting the requirements set forth in this request for bids.
- 4) Description of vendor's internal QA/QC procedures, which will insure the highest level of accuracy in both the sorting and identifying processes.
- 5) Actual bid – see 'Bid Schedule'.

## SPECIFICATIONS FOR PERIPHYTON PROCESSING AND IDENTIFICATION

### “Soft” (Non-Diatom) Algae – Relative Abundance and Taxa Richness

Homogenize the sample with a blender. Pipette a subsample into a Palmer counting cell. Permanent mounting techniques can be utilized if preferred. Dilute samples if cells overlap too much for counting. Identify and count 300 algal non-diatom units to the lowest taxonomic level at magnification of at least 400X (OK to use additional levels of magnification). Cell units, of 10  $\mu\text{m}$  length, should be counted instead of individual cells for filamentous species (or measure average cells per filament based on average cell length per filament). Individual cells of colonial species should be counted when appropriate. Count live and dead (those with no cell content) diatoms separately, recording only the number of each observed in order to determine live: dead diatom ratio (identification will be done on the cleaned samples). Record numbers of non-diatom algal units on the non-diatom bench sheet that should be similar to the example provided in Appendix A of EPA's *Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers 1999* (Barbour et. al.) [http://www.epa.gov/owow/monitoring/rbp/app\\_a.html](http://www.epa.gov/owow/monitoring/rbp/app_a.html).

### Calculate Biovolume / Biomass

Biomass should be calculated for the most abundant taxa (> 10% of sample) at minimum, it is fine to include measurements for all taxa. A minimum of 15 measurements (length and width) should be taken for these species. The average measurements are used to calculate the biovolume, which is then converted to biomass, assuming a specific gravity of 1. The biomass of each species is calculated based on the abundance of that species and adjusted for original sample volume.

### Diatoms

After the non-diatom algae have been identified, clear diatom frustules of organic and intercellular material using either ‘Nitric Acid Oxidation’ or ‘Hydrogen Peroxide / Potassium Dichromate Oxidation’.

Prepare slides and identify diatoms at 1000X to the lowest possible taxonomic level, preferably to the species or variety level, using current taxonomic references. Record all taxa encountered on the diatom bench sheet creating a species list prior to enumeration. Scan the slide until several minutes pass without producing any new taxa. For quantitative data, count a minimum of 600 valves recording taxa and number counted on the diatom bench sheet. Diatom biovolume should be determined using the NAPHRAX mounts.



**Reporting**

All taxa, counts, and biomass information should be entered into an electronic format (either database or spreadsheet – the format should be discussed with DWWM prior to populating with data) with all site information, including stream name, stream code, stream mile (if provided), date collected, collector's name, taxonomist's name, and date identified. If vendor typically calculates metrics, provide bids both with and without these calculations. Other information to be submitted must include: Method for clearing diatoms, procedure used for enumerating colonial and filamentous algal species.

**BID SCHEDULE**

Bid:

\$ \_\_\_\_\_ Per sample to process and identify samples – including cleaning and preparation of slides, and adherence to all QA/QC procedures outlined in bid proposal.

Contact Person: \_\_\_\_\_

Contact Phone #: \_\_\_\_\_

**AFFIDAVIT****West Virginia Code §5A-3-10a states:**

No contract or renewal of any contract may be awarded by the state or any of its political subdivisions to any vendor or prospective vendor when the vendor or prospective vendor or a related party to the vendor or prospective vendor is a debtor and the debt owned is an amount greater than one thousand dollars in the aggregate.

**Definitions:**

"Debt" means any assessment, premium, penalty, fine, tax or other amount of money owed to the state or any of its political subdivisions because of a judgment, fine, permit violation, license assessment, defaulted workers' compensation premium, penalty or other assessment presently delinquent or due and required to be paid to the state or any of its political subdivisions, including any interest or additional penalties accrued thereon.

"Debtor" means any individual, corporation, partnership, association, limited liability company or any other form or business association owing a debt to the state or any of its political subdivisions.

"Political subdivision" means any county commission; municipality; county board of education; any instrumentality established by a county or municipality; any separate corporation or instrumentality established by one or more counties or municipalities, as permitted by law; or any public body charged by law with the performance of a government function or whose jurisdiction is coextensive with one or more counties or municipalities.

"Related party" means a party, whether an individual, corporation, partnership, association, limited liability company or any other form or business association or other entity whatsoever, related to any vendor by blood, marriage, ownership or contract through which the party has a relationship of ownership or other interest with the vendor so that the party will actually or by effect receive or control a portion of the benefit, profit or other consideration from performance of a vendor contract with the party receiving an amount that meets or exceed five percent of the total contract amount.

**Exception:**

The prohibition of this section does not apply where a vendor has contested any tax administered pursuant to chapter eleven of this code, workers' compensation premium, permit fee or environmental fee or assessment and the matter has not become final or where the vendor has entered into a payment plan or agreement and the vendor is not in default of any of the provisions of such plan or agreement.

Under penalty of law for false swearing (West Virginia Code, §61-5-3), it is hereby certified that the bidder and all related parties do not owe any debts or, if a debt is owed, that the provisions of the exception clause above apply; and all state licensing requirements are in compliance.

Vendor's Name: \_\_\_\_\_

Authorized Signature: \_\_\_\_\_ Date: \_\_\_\_\_