Maryland Biological Stream Survey

Round Four Sampling Manual

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DEPARTMENT OF
NATURAL RESOURCES

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Maryland Biological Stream Survey: Round Four Field Sampling Manual

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Foreword

This document titled "The Maryland Biological Stream Survey: Round Four Sampling Manual" was prepared by staff in the Maryland Department of Natural Resources (DNR), Monitoring and Non-Tidal Assessment Division, with input from Versar Inc. and the University of Maryland Appalachian Laboratory staff. It provides written standard operating procedures for all aspects of the Round Four Maryland Biological Stream Survey (MBSS) sampling. The primary users of this manual are DNR and University of Maryland field crews collecting data as part of Round Four (2014 – 2018) of the MBSS. Additionally, to facilitate data comparability and sharing, we encourage the use of this manual by anyone collecting stream ecological data during Round Four, especially those seeking certification in certain aspects of MBSS benthic macroinvertebrate or fish sampling protocols.

Purpose of Manual

This document was prepared to support the Round Four MBSS. It is imperative that the protocols used for every aspect of the MBSS be provided to help guarantee the collection of consistently high-quality data throughout Round Four and to ensure that the goals and objectives of the Round are met. These written protocols also provide information to anyone attempting to duplicate procedures used by the MBSS and to ensure comparability of data and results generated by the MBSS. All persons working on the MBSS or generating reports using MBSS protocols should be familiar with the information provided herein. Certification in certain MBSS sampling protocols was first offered in 2012. This manual describes protocols for three aspects of MBSS sampling for which certification is offered, including Benthic Macroinvertebrate Sampling, Fish Crew Leader, and Fish Sampling. This manual should be the primary reference for detailed descriptions of these protocols. The manual also provides detailed descriptions of all other field sampling protocols to be followed by DNR MANTA staff and anyone else sampling as part of the Round Four MBSS. Additional information regarding the MBSS certification process, other aspects of the MBSS for which certification is being offered, and requirements for attaining certification can be found on this Maryland Department of Natural Resources web site (www.dnr.maryland.gov/streams/Pages/mbsstraining.aspx).

Maryland Biological Stream Survey Round Four Goal and Objectives

The original goal and objectives of the MBSS from previous rounds are still germane to Round Four. The goal is to provide the best possible information for ensuring the protection and restoration of Maryland's stream ecological resources. There are four objectives that the MBSS pursues to attain this goal: 1) Assess, with known confidence, the current condition of ecological resources in Maryland's streams and rivers; 2) Identify causes of adverse effects (stressors) to ecological resources; 3) Provide an inventory of biodiversity in Maryland's streams; and 4) Document changes (improvements and degradation) over time in Maryland's stream ecological conditions and biodiversity status.

The fourth objective – "document changes (improvements and degradation) over time in Maryland's stream ecological conditions and biodiversity status" - is the primary focus of Round Four. To optimize the chance for detecting changes, the Round Four design consists of re-sampling sites that were randomly-selected and sampled during Rounds One (20 years later) and Two (14 years later). More details regarding the Round Four sampling design are available in Appendix A.

Round One (1995 – 1997) provided Maryland's first statewide assessment of non-tidal stream ecological conditions (Objective 1). The information from Rounds One and Two was also useful in identifying many of the most pervasive stressors (Objective 2) and providing a preliminary inventory of Maryland's stream biodiversity (Objective 3). Although changes in ecological conditions (Objective 4) between the first two rounds were examined, different stream maps were used as a basis for selecting sites during these two rounds. Thus, the data generated from these different maps are not directly comparable for detecting changes in stream conditions. The same stream map was used during Rounds Two (2000-2004) and Three (2007-2009). Data generated from randomly-selected sites sampled during these two Rounds, over approximately a seven year interval, did

not show significant differences in estimated statewide stream conditions. The Round Four design optimizes the ability to compare stream conditions over time by sampling the same sites (thus decreasing the variability introduced by comparing different sets of randomly-selected sites) that were sampled previously and compares conditions over the longest possible time intervals (20 and 14 years). Although the condition of Maryland's individual 8-digit watersheds will not be provided from Round Four, statewide and basin (6-digit Maryland watershed) assessments of stream ecological conditions will be available and can be compared to results from Rounds One and Two.

Introduction

Data and results generated by the MBSS have been widely used for making management decisions. Examples include Maryland's 305b report to Congress and the list of impaired waters (303d list), as well as identification of Tiered Aquatic Life Uses and aquatic biodiversity priority areas. Additionally, data from the long-term monitoring of reference (Sentinel) sites in Maryland are being used to explore potential natural variability and variability in stream ecology that may be attributable to climate change. The quality, usefulness, and availability of MBSS data are also exemplified by their use in a large number of peer-reviewed scientific publications. We anticipate the Round Four data being useful for examining potential changes in Maryland's stream conditions over time.

To achieve this objective, examine for potential changes over time, it will be important to be able to compare data collected during Round Four with data from Rounds One and Two. Thus, it is important to make sure that the protocols used for collecting data between Rounds are as similar as possible. The protocols for collecting benthic macroinvertebrate and fish data in the field have not changed since the first year of the MBSS (1995). The only change has been to eliminate the examination of each fish collected for external anomalies after the completion of Round One. During Round Four, fish will be examined for external anomalies at sites that were previously sampled during Round One so that anomaly data can be compared over the 20 year period between Rounds One and Four.

Protocols for collecting water chemistry samples have also remained largely consistent since 1995 and laboratory methods have not changed at all. During Round One, six water chemistry variables (pH, ANC, sulfate, nitrate, conductivity, and DOC) were measured from spring water grab samples. At the beginning of Round Two (2000), six parameters (chloride, total nitrogen, nitrite, ammonia, total phosphorus, and orthophosphate) were added to provide better assessments of nutrient concentrations. At the beginning of Round Three, in situ summer measurements of pH, temperature, conductivity, and dissolved oxygen were no longer being taken because all of these (with the exception of dissolved oxygen) are available from the spring water sample or temperature loggers. The water chemistry sampling for Round Four will follow the Round Three procedure, with the addition of copper, zinc, magnesium and calcium.

Most physical habitat assessment measurements and assessments have remained consistent since the first year of the MBSS. However, a few procedures were added at the beginning of Round Two and a few were modified. During Round Four, we will continue to use the methods employed at the beginning of Round Two. However, we will add several assessments conducted using Round One protocols to the repeated Round One sites. These include assessments of bank stability, minimum buffer width, channel flow status, and remoteness. In Round Four, the "Presence, Absence, or Extensive Presence of orange floc" was added to the "Stream Character" portion of the MBSS Summer Habitat Data Sheet. Along with the physical habitat assessment, during Round Four, geomorphological measurements will be taken at MBSS sites. These measurements will consist of a cross section, pebble count, and facies mapping.

Freshwater mussel, crayfish, and stream salamander data collection differed by MBSS Round. Round Four methods for sampling freshwater mussel and crayfish will be the same as the methods used during Round Three. However, additional effort will be employed for detecting and quantifying stream salamander abundance during Round Four. This extra effort is deemed necessary for testing the use of stream salamanders in a stream salamander index of biotic integrity.

The MBSS Round Four protocols are designed to optimize comparability with Rounds One and Two and add important information for assessing Maryland's streams into the future. Detailed protocol descriptions are provided in the remainder of this document. Persons conducting MBSS Round Four sampling should follow these descriptions and have this document with them while conducting Round Four sampling.

1.0 Personnel and Crew Qualifications

Persons responsible for field collection of MBSS data fit into one of three positions: Crew Supervisor, Crew Leader, or Crew Member. Each position is responsible for different aspects of field data collection. The specific responsibilities of these positions as they apply to each aspect of MBSS field data collection are described, along with the description of each aspect. Specific qualifications for each MBSS position are as follows. The Crew Supervisor must be familiar with all aspects of MBSS sampling and have a minimum of five years of experience leading field data collection efforts and the logistics involved with planning and implementing field data collection. The Crew Leader must be intimately familiar with every aspect of MBSS sampling and have at least three years of experience with MBSS sampling or with another comparable ecological field sampling effort. Crew Member qualifications are less stringent; however persons in this position must be physically fit for strenuous activity and must follow all safety, data collection, and quality control procedures.

Along with having qualified persons in each of these positions, all MBSS field Crew Members and the Crew Leader must have received training in MBSS protocols during each year they are field Crew Members or a Crew Leader. Additionally, the field crew must be made up of persons who collectively passed all MBSS taxonomy tests for any taxonomic groups on which the crew plans to collect data and identify live organisms in the field. For example, the fish taxonomy test must be passed by at least one person on the crew to identify fishes, and that person must conduct the fish taxonomic identifications. Since benthic macroinvertebrates are identified in the laboratory, no one on the MBSS field crew is required to pass the benthic macroinvertebrate taxonomy test to collect benthic macroinvertebrates.

To ensure comparability among data collected by different sampling crews, all crews must also commit to regular field audits (see chapter 3.5, Quality Assurance). Typically, audits are performed at a minimum of two sites sampled by each crew by the MBSS QC Officer. However, additional audits may be required depending on the experience of the crew, performance on previous audits, and intended use of collected data. Audits can only be performed by a qualified MBSS QC Officer. This individual has had extensive MBSS crew leader experience, has extensive experience in conducting MBSS training, and is familiar with the intended use of MBSS data by the crew being audited. The QC Officer should also be familiar with aspects of the MBSS other than field data collection (e.g., laboratory protocols, IBI calculation, data management).

2.0 Health and Safety

The purpose of this chapter is to provide recommendations for health and safety aspects to persons involved in MBSS field collections. Suggested training and qualifications are described, along with general safety procedures, sampling hazards, provision of first aid, and emergency situations. The ultimate responsibility for health and safety of field crews lies with the parent organization for each field crew.

2.1 Training and Qualifications

To minimize any potential health and safety risks related to field sampling conducted as part of the MBSS, survey personnel need to be physically able to conduct fieldwork under demanding conditions and be well prepared to handle contingencies or emergencies. The following are suggested requirements for all field survey personnel:

- Recent (within 2 year) physician's approval to conduct rigorous physical work
- Recent (within 2 year) CPR certification
- Recent (within 2 year) Red Cross First Aid Training
- Complete a satisfactory interview about health and safety aspects of the MBSS with the Field Crew Supervisor, including routine safety precautions and a discussion of actions to be taken in an emergency.

In addition to the recommendations identified for all survey personnel, Crew Leaders should have adequate field sampling experience under rigorous conditions.

2.2 Duties and Responsibilities

This section outlines the health and safety responsibilities of persons involved with MBSS field activities.

2.2.1 Field Crew Supervisor

The Field Crew Supervisor for each organization involved in stream sampling has overall responsibility for health and safety aspects of the portion of the MBSS for which that organization is responsible.

2.2.2 Crew Leader

Field Crew Leaders are responsible for ensuring that day-to-day activities of the field crew are conducted in a safe manner. Recommended health and safety responsibilities of the Crew Leader include:

- Instructing and supervising the survey crew such that sampling at and travel at a given site are done in a manner which minimizes health and safety risks;
- Reporting to the Field Crew Supervisor or his/her designee any unusual health and safety conditions, emergencies, or accidents encountered during the deployment of the crew. In the case of accidents or emergencies, the Crew Leader should, as soon as the situation permits, notify the Field Crew Supervisor or his/her designee by direct phone contact;
- Ensuring that vehicles and sampling equipment are in safe operating condition prior to and during field deployments;
- Ensuring that all members of the survey team are fully aware of any potentially hazardous materials used as part of sampling. Examples include preservatives for biological and chemical samples;
- Determining whether sampling conditions are safe and appropriate;
- Informing the survey team of any situation-specific dangers involved at a given site;
- Ensuring that vehicles are operated in a safe manner; and
- Ensuring that samples and sampling equipment are safely stored prior to vehicle operations.

2.2.3 Field Crew Members

All personnel involved in field sampling or field observations (e.g., QA/QC inspections) should be aware of the risks involved with the routine aspects of MBSS. When unsafe or hazardous conditions are observed, crew members should inform the Crew Leader at the earliest opportunity. In addition, crew members should notify the Crew Leader if, for any reason, they cannot perform an assigned task in a safe manner. Examples include sickness, physical limitations, or uncertainty about proper operation of the sampling equipment. Field crew members should also inform the Crew Leader of any allergies or medical conditions (e.g., diabetes, asthma, allergies) and any special needs (e.g., inhaler, epinephrine pen) the crew member has. It is the responsibility of each crew member (not the Crew Leader) to make sure he or she has any special needs medicine or equipment and that the Crew Leader knows about that special need.

2.3 Sampling Hazards and Procedures for Minimizing Risk

There are a number of potential health and safety considerations specific to the MBSS. A number of these hazards are common to all sampling sites, while others may be site- or region-specific. This section lists a

number of hazards likely to be encountered during the MBSS as well as measures to minimize the health and safety risks associated with them.

1. **Vehicle Accident.** As with nearly all other field sampling programs, there is a risk of a vehicular accident. To minimize this risk, the following measures should be taken:

   - An inspection of the sampling vehicle should be performed by the Crew Leader or a designee prior to sampling departure. This inspection should include tire condition and operability of wipers, defroster, etc.;

   - During sampling activities, any potentially unsafe vehicle condition should be reported to the Field Crew Supervisor and corrected as soon as is practical;

   - If, in the judgment of the Crew Leader, the sampling vehicle is not safe to operate, the vehicle should not be operated until the condition is rectified;

   - Vehicles should not be operated by crew members who are incapable of safely operating them. No sampling vehicle should be operated by a person not holding a valid drivers license.

2. **Electric Shock.** Failure to observe appropriate safety precautions when using backpack electrofishing gear could result in electric shock. Under worst case conditions, this shock could result in cardiac arrest and loss of life. To minimize risks associated with electrofishing during the MBSS, the following measures shall be taken:

   - Only personnel designated by Field Crew Leaders should operate the backpack electrofishing unit;

   - To minimize the amount of body surface area potentially exposed to electric shock, normal wading gear for the MBSS should be chest waders. Only non-leaking wading gear should be used during electrofishing-- if a leak is discovered, wading gear should be changed and the leaking gear repaired or replaced prior to the next use;

   - Bare wire portion of the cathode (rattail) or the anode should never be touched while the unit is in operation;

   - Electrofishing should only be conducted when a minimum of three persons are present at a site. In the event of electric shock, this provides for one person to administer CPR while another seeks medical assistance. Use of a portable phone is also recommended as an effective means to summon emergency medical care if necessary;

   - If the Crew Leader determines that stream conditions at the time of the site visit present an abnormal risk of electric shock, he or she will determine that the site is not sampleable and sampling will be conducted at an alternate site or canceled in that reach;

   - Prior to each use, electrofishing gear should be verified to be in safe working condition by the Crew Leader. This verification should include an examination of external wiring and electrical connections; and

   - In cases where two electrofishing units are used or barge shocking is employed at a site, extra care should be taken to ensure that unit operators maintain an awareness of all personnel in the water. In addition, unit operators should maintain adequate spacing between units to minimize the risks of shock from both electric fields in the event a crew member slips or falls into the water, or the discharge of one anode completing the switch circuit for another unit.

3. **Hazardous Terrain.** A routine part of sampling during the MBSS is traveling over rough terrain to access the sample site. One of the risks arising from this aspect of the MBSS is the possibility of injury from falling. To minimize this risk, the following preventive actions are recommended:

   - When necessary, the Crew Leader will make a determination that access to the sampling site is not possible and the site will be deemed unsampleable;
   - When traveling over any extensive distance, appropriate footwear should be worn instead of waders or hip boots;
   - Equipment should be distributed equitably among crew members for transport from the vehicle to the site. If determined to be necessary by the Crew Leader, more than one trip to transport equipment should be made;
   - To the greatest extent possible, travel between the vehicle and the sample site should occur during daylight hours; and
   - Only in unusual circumstances (as determined by the Crew Leader) should a crew member travel alone over hazardous terrain.

4. **Fast or Deep Water.** During the MBSS, some sampling sites may be visited which contain fast and/or deep water in them. Sampling in locations which are too deep or too fast for wading could result in injury or drowning. It should be noted that sampling fast and/or deep waters also increases the likelihood of electrical shock; thus a high degree of caution is imperative for safe operations. To minimize health and safety risks associated with sampling in fast and/or deep waters, the following steps should be taken:

   - Prior to sampling, the Crew Leader should ensure that all crew members who are to enter the stream are physically fit to do so and are aware of any specific sampling risks at the site;
   - Prior to sampling, the Crew Leader should make a determination as to whether the site can be sampled by wading without undue risks. If a negative determination is reached, the site should be revisited at another time or not sampled;
   - Field Crew Members should wear chest waders outfitted with waist belts and sticky rubber soled wading boots and/or cleats should be used in rocky areas. Felt soled boots are banned throughout the State of Maryland and should not be worn to avoid the transmission of harmful aquatic organisms.

5. **Slippery Substrate.** During the MBSS, sampling at some sites will be hazardous due to slippery substrate. Examples of stream types which may have treacherous substrates include those affected by acid mine drainage and streams with high silt loads. To minimize the risks associated with slippery substrates, the following measures are recommended:

   - The Crew Leader should factor the slipperiness of the substrate into decisions as to whether a site can be sampled and any extra precautions to be taken by the field crew; all wading gear should have sticky rubber soled wading boots and/or cleats.

6. **Dangerous Animals or Plants.** Sampling at some MBSS sites will include risks associated with dangerous animals and/or plants. Poison ivy is likely to be common along many travel routes used by the sampling crew, as well as in riparian vegetation. Poison ivy roots on tree trunks offer particular risks since they are often unnoticed. Poison sumac, which occurs in boggy areas, should also be avoided. Contact with bees, wasps, and certain caterpillars can cause allergic reactions and should also be avoided. A number of other animals also present serious risks including: northern copperheads, timber rattlesnakes, free-ranging

domestic dogs, rabid animals of any species, and ticks. To minimize the risks associated with dangerous animals and plants during the MBSS, the following measures are recommended:

- All field survey personnel should receive training in field identification, avoidance of, and first aid for dangerous plants and animals which may be encountered during the MBSS;
- Crew members should inform their Crew Leader of any known allergies and keep appropriate medical relief in the first aid kit (at a minimum, each crew should keep an emergency supply of Benadryl – gel caps or liquid are preferred because they enter the bloodstream more quickly than tablet form);
- The Crew Leader should make all crew members aware of site- or situation-specific dangers. Similarly, field crew members should inform the Crew Leader as soon as they are discovered;
- All crew members should be informed of the risks of Lyme disease and should check themselves after conducting field work for ticks that may have become attached to the body.

7. **High Bacterial Levels.** When sampling in areas downstream of sewage or other organic waste sources, potentially dangerous bacterial levels may exist. In urban areas, the presence of such materials may be clearly evident by smell, observation of solids and floatables, and/or the presence of sewage fungus on bottom substrates. However, in some areas, potentially dangerous bacterial levels could be present in a stream without any obvious evidence. To minimize the health risks associated with high bacterial levels in streams, the following measures should be incorporated into field surveys:

   - During development of the itinerary, the Crew Leader should examine the list of NPDES discharge permits and investigate through MDE any known pollution problems in the watershed being sampled. Using this information, a determination should be made as to whether special safety precautions are necessary;
   - Prior to entering the stream, the Crew Leader should make note of any evidence of high bacterial levels and inform the field crew;
   - The use of gloves should be maximized during the sampling process;
   - Open wounds should not be exposed to contact with stream water; and
   - After exposure to stream water, all crew members should wash their hands in isopropyl alcohol and clean water prior to consuming any food or drink.

8. **Hazardous Waste.** Because of historical disposal practices, hazardous wastes may be present at an unknown number of sites to be sampled during the MBSS. Risks of relatively brief exposure (such as sampling during the MBSS) to hazardous wastes are likely to be low, but precautions still need to be taken to minimize exposure probabilities. These include:

   - Prior to commencement of field sampling, existing information (through MDE and EPA) about known or probable hazardous waste sites in Maryland in relation to MBSS sample sites should be reviewed. After review of available hazardous site information, the crew should be informed of any hazardous waste sites in areas designated to be sampled. Any such areas identified will be sampled by a crew that has received OSHA hazardous waste safety training (as specified in 29 CFR 1910.120);
   - All sampling at hazardous waste sites will be conducted in accordance with site health and safety plans and only after proper advance notice has been given to authorities on site;

- If sampling is to be conducted in an area where hazardous waste is known to be present, MBSS personnel who participate in sampling should participate in a Medical Monitoring Plan established by the Contractor for the hazardous site sampling crew. Medical Monitoring should include baseline, yearly, and exit examinations;

- After sampling at or in the vicinity of hazardous waste sites, all exposed equipment should be thoroughly rinsed, including waders and any exposed personal equipment and;

- No food should be consumed at known hazardous waste sites and following sampling, food will only be consumed after thorough hand washing.

9. **Hypothermia.** Many of the sites sampled during the MBSS will be in remote locations. At these locations, the potential for stranding and prolonged exposure to extreme weather conditions is of concern, especially when sampling is conducted during cold weather. There is also a potential for prolonged exposure to cold water in the case of accidents, emergencies or other unusual conditions. Recommended precautions to reduce the possibility of hypothermia or related illnesses include:

    - Each field crew should carry several space blankets at all times when in the field during the Spring Index Period;

    - Crew Leaders should be responsible for monitoring weather conditions and adjusting or postponing sampling plans as appropriate; and

    - Prior to leaving the vehicle for a sampling site, the Crew Leader should ensure that crew members are properly clothed and that emergency supplies are taken to the site.

10. **Lightning Strike.** As sampling during the MBSS will occur over relatively long periods of time in spring and summer, exposure of field crews to electrical storms is likely. To minimize risks associated with a lightning strike the following measures should be taken:

    - Crew Leaders should be responsible for monitoring weather conditions, adjusting sampling schedules as appropriate to minimize the chance of a field crew being exposed to an electrical storm while in a remote location; and

    - In the event of an electrical storm while sampling, sampling activities should be halted and the Crew Leader should determine whether to return to the vehicle or seek local shelter.

11. **Dehydration and Hyperthermia.** The most prevalent risk to MBSS sampling crews is the risk of dehydration. Freshwater should be kept with sampling crews at all times and crew members should be encouraged to drink plenty of water. In the event that a crew member suffers from dehydration or heat related illness, all possible attempts should be made to cool and hydrate the person. Make sure to have plenty of fresh drinking water readily available.

2.4 First Aid

During any field sampling activity such as the MBSS, there is a possibility that first aid will need to be administered. To meet this need, all personnel should be trained in first aid. In addition, each field crew should maintain a stocked first aid kit in both field sampling equipment and in the sampling vehicle.

2.5 Emergencies

In the event of a medical or other emergency, the Crew Leader or qualified crew member should take all appropriate immediate actions and should send for appropriate assistance using the fastest available means. In

the event the emergency occurs at a remote location, all necessary information to guide assistance personnel should be provided, including map coordinates if known and appropriate.

2.6 Precautions for Minimizing Ecological Risk (Decontamination)

An increasing potential exists for transferring non-native and invasive organisms (including those that cause serious diseases to native stream dwelling fauna and fauna) from one stream to another while conducting monitoring. Whirling disease (a protist, *Myxobolus cerebralis*), Didymo (an algae, *Didymospenia geminata*), and amphibian chytrid fungus (*Batrachochytrium dendrobatidis*) are examples of such organisms. In addition, avian influenza can be transferred among farms simply by walking in the poultry litter that came from infected poultry and then walking in another area with poultry. It is important to properly clean all footwear or other equipment that may have contacted disease-containing litter.

The risks described above require that field crews conducting MBSS sampling take precautions to minimize, to the greatest extent possible, the transfer of any disease organisms from one place to another. Since 2007, all MBSS field crews have been required to disinfect all field equipment and waders that come in contact with stream or wetland (e.g. vernal pool) water following sampling at each stream site. This procedure should also be applied to all equipment that comes in contact with poultry litter.

The disinfection procedure consists of soaking or rinsing all equipment that has come in contact with water (or poultry litter) in a 10% bleach or 2% Virkon solution for at least one minute. Equipment with a smooth surface (e.g. buckets, sides – but not soles - of waders) can sprayed with bleach or Virkon solution. After soaking and scrubbing have been completed, all equipment must be rinsed with freshwater to remove the bleach or Virkon solution. Avoid skin and eye contact with bleach solution as it can be severely irritating. Thoroughly rinsing all equipment with freshwater also minimizes risk of skin and eye irritation. Decontamination should occur away from surface waters to avoid polluting them.

3.0 Quality Assurance

The purpose of this chapter is to outline QA/QC activities that are part of MBSS field activities. The chapter includes descriptions of documentation procedures, responsibility and accountability of project personnel, training requirements, facilities, and equipment. To achieve the objectives of the MBSS, it is imperative that all project personnel follow the procedures and guidance provided in this chapter.

3.1 Introduction

Quality assurance and quality control (QA/QC) are integral parts of data collection and management activities of the MBSS. The field QA program for the MBSS was designed to: 1) ensure comparability of data collected by sampling crews and to data collected previously by the MBSS, 2) ensure that data are of known and sufficient quality to meet the project objectives, and 3) provide estimates of various sources of variance associated with the individual variables/parameters being measured.

To be effective, the QA program must continually monitor the accuracy, precision, completeness, and comparability of the data during all phases of the program. Components of the MBSS field QA program include:

- thorough and annual training of all persons involved with data collection;
- development of and adherence to strict project protocols and guidelines;
- comprehensive field and laboratory data documentation and management;
- verification of data reproducibility; and

- proper calibration of all equipment used for data collection.

3.2 Stream Population of Interest

The current population of interest for the MBSS includes all non-tidal, 4th order and smaller stream reaches of the State of Maryland, with the exception of reservoir-like impoundments which substantially alter the lotic nature of the reach.

3.3 Comparability and Completeness

Comparability of data between field crews is maximized by providing standardized training in MBSS techniques prior to each sampling period. Training requirements are included in the Scope of Work for each organization involved in field sampling. Training is mandatory for all persons involved with MBSS data collection.

To utilize data from a given site during analyses, all data included on the MBSS data sheets, which pertains to the analysis being conducted, must be validated along with appropriate site location information.

3.4 Documentation

To ensure scientific credibility, study repeatability and cost effectiveness, all field sampling activities of the MBSS need to be adequately documented. These activities include adherence to sampling protocols, equipment calibration, data sheet review, field notes, information management, and data quality assessment. To minimize the possibility that needed documentation or data are not recorded, standardized forms and on-site verification of form completions by supervisory personnel are included as part of the MBSS. Each of the activities listed above is described in other sections of this manual, including documentation procedures and requirements.

3.5 Field Audits

For the field data collection component of the MBSS, the QC Officer is primarily responsible for conducting field audits. At least one site sampled by each MBSS crew during each index period should be subject to audit. However, additional audits may be required depending on the experience of the crew, performance on previous audits, and intended use of collected data. Field audits consist of checking for consistency and accuracy in taxonomic identification, site location confirmation, calibration and maintenance of equipment, adherence to established protocols, record keeping, and prompt identification of necessary remedial or corrective actions.

For taxonomic identification, the QC Officer may designate someone who is an expert in a particular taxonomic group to verify accurate taxonomic identification.

To ensure consistency in data collection, the QC Officer is required to fill out an extra set of MBSS data sheets at sites sampled during field audits. These data sheets are to be filled out independently from the data sheets filled out by the crew. Any decisions regarding safety, sampleability, number of persons involved with sampling at the site, use of equipment, or anything that may affect data quality, comparability, or completeness should be recorded on the extra data sheets or in a QC log book. The data recorded by the QC Officer will be compared to the data recorded by each crew. Assuming the QC Officer makes decisions and records data consistently, and since the QC Officer visits all sampling crews, this provides a measure of comparability of data collection among sampling crews. In addition to field audits, the QC Officer will visit with each crew prior to the Summer Index Period to verify competency prior to initiating sampling. This visit typically consists of protocol review in the field while hypothetical sampling is conducted.

3.6 Training Requirements

An important aspect of the MBSS QA program is the training program for field personnel, which will be conducted prior to each spring and summer sampling period. Training helps to ensure consistent implementation of required procedures and attainment by each person of a minimum level of technical competency. **All**

participants in MBSS field sampling must receive MBSS training. Additionally, the field crew must be made up of persons who collectively passed all MBSS taxonomy tests for any taxonomic groups on which the crew plans to collect field data and identify organisms to species in the field (e.g., at least one member of each field crew must pass the fish taxonomy test every year to be qualified to collect MBSS fish data). See section 3.3.11 for details regarding taxonomic identification as it pertains to MBSS field sampling. Since benthic macroinvertebrates are identified in the laboratory, no one on a MBSS field crew is required to pass a benthic macroinvertebrate taxonomy test to collect benthic macroinvertebrates.

3.7 Equipment Maintenance and Calibration

Preventive maintenance and calibration must be performed on all sampling equipment used as part of the MBSS. Maintenance and calibration procedures should be implemented as per manufacturer instructions. Unless otherwise specified, calibration must be performed daily prior to equipment use and anytime equipment problems are suspected. Preventative maintenance must be performed at intervals not to exceed the frequency recommended by the manufacturer. All equipment malfunctions must be fully corrected prior to next use. For weighing scales, weekly checks must be conducted during field sampling using NIST standards or other accepted standards to demonstrate that instrument error is within limits specified by the manufacturer.

For each item of equipment used as part of the MBSS, a bound logbook for calibration and maintenance must be maintained. Entries in the log must be made for all calibration and maintenance activities. Documentation includes detailed descriptions of all calibrations, adjustments, and replacement of parts, and each entry must be signed and dated.

To ensure that MBSS equipment is operated within QA/QC requirements, the QC Officer should conduct periodic site equipment audits.

3.8 Field Information Management

Each MBSS site is assigned a unique identification code. The code is recorded at the top of all MBSS data sheets. The unique code is made up of four parts. 1) Watershed code. The appropriate four letter code indicating the eight digit watershed containing the site (watershed codes are found in Appendix D). 2) Segment. Three numbers are used to designate the stream segment obtained from the appropriate reach file. These three letters begin with the stream order and the next two letters refer to the order in which the site was selected. For random sites, the order in which the sites were collected can be important as sites lower in order being sampled indicate less probability of bias (i.e. in being representative of watershed conditions) compared to having many sites with higher order sampled. 3) Type. A one letter code is used to designate the site type. Examples of site type codes include "R" for random sites, "S" for sentinel sites, "X" for special study sites and "T" for targeted sites. 4) Year. The last four digits in the site identification are the calendar year during which sampling occurred.

To facilitate data recording during inclement weather, data sheets should be printed on waterproof paper. Backup copies of all field data sheets must be made. Digital photographs should be labeled appropriately with site identification and backed up.

To ensure that all field data for the MBSS are collected and recorded in a usable manner, all data should be printed in the units specified on the MBSS data sheets. No writing over is permitted on data sheets. The incorrect entry should be lined out and the correct entry written in an obvious location next to the line out. Data sheets for a given site must be consecutively labeled so that the total number of data sheets generated for each site is known. Recorded data must be reviewed at the point of entry and the Crew Leader and one other member of the crew must review and initial all data sheets prior to departure from the site.

Each sample collected as part of the MBSS will be assigned a sample number. The sample number will contain several unique identifiers to minimize the possibility of misidentification. In addition, chain-of-custody

forms should be maintained for all water and benthic macroinvertebrate samples (Appendix G), as well as herpetofauna, crayfish, mussel, and fish voucher specimens.

3.9 Data Quality Assessment

Assessment of data quality against established data quality objectives will be conducted to determine the overall performance of the QA program, identify potential limitations to use and interpretation of the field collected data, and to provide information for other data users regarding usability of the data for other purposes.

The quality of MBSS data will be evaluated in several ways. Precision and bias associated with important elements of the sampling and measurement process for each variable measured will be evaluated using results from replicate sampling and performance evaluation studies. Information about precision, bias, and completeness will be used to determine the comparability of data acquired during each sampling year.

Inherent differences in data collected at individual sites are potentially confounded by crew differences in sampling efficiency, experience, knowledge of protocols, or sampling effort. Such crew differences can adversely affect data quality and interpretation of regional patterns, but logistics constrain the degree to which these potential limitations can be evaluated and/or corrected. In general, field crews will be assigned sampling sites within discrete geographic regions, and it is likely that sampling efficiency will not be uniform from the beginning to the end of the index period or between years. To minimize this effect, retaining consistent personnel should be a priority.

3.10 Duplicate Samples

To aid evaluation of precision and bias, 5% of all MBSS sites will have replicate benthic macroinvertebrate and water chemistry samples collected. For water chemistry samples, one QC sample from each crew will be a blank (filled with deionized water); the remainder of the 5% will be duplicates. These samples are in addition to other duplicate and blank samples analyzed as part of in-laboratory QA/QC protocols. An annual summary of QA/QC results for benthic macroinvertebrate and analytical chemistry sampling will be prepared and maintained on file.

3.11 Taxonomic Identification and Specimen Vouchering

The MBSS is recognized as providing the highest quality biological data. This is due primarily to the QA/QC requirements for taxonomic identification. The following taxa are identified to species (or sub-species in some cases) in the field: fishes, reptiles, amphibians, crayfishes, freshwater mussels, and select invasive plants. The crew conducting MBSS sampling must consist of members who, collectively, have passed identification tests for all of these taxonomic groups. Only the person(s) on each field crew who has passed the test for the taxonomic group should conduct identification in the field.

During the Round Four MBSS, each field crew should maintain a voucher collection of at least five specimens of each fish, herpetofauna, mussel, and crayfish species encountered (as long as five were collected). The voucher collection can consist of photographs and/or preserved specimens of each species. Photographic vouchers will be accepted in lieu of preserved specimens, as long as the diagnostic features for identifying the specimens photographed can be seen clearly. Dead mussel shells should be retained. Live mussels should be photographed. Specimens of crayfishes should be preserved in lieu of photographs when possible. Any rare, threatened, or endangered (see Appendix F) species encountered should be photographed and not preserved. Nuisance species (Appendix F) should be photographed or preserved. Care should be taken while photographing to avoid harm to the specimen. Photographs must clearly show the appropriate features necessary for identifying the species. With the exception of rare, threatened, or endangered species, specimens that are too small to provide photographs that can be used to verify identifications (or with diagnostic features that do not show up well in photo-

graphs) should be preserved for verification. Please see Appendix B for detailed fish fixation and preservation procedures.

To facilitate record keeping of vouchered specimens and QA/QC verification of species identifications, each MBSS field crew leader should maintain a list of all specimens vouchered (photographed or preserved) during the MBSS Spring and Summer Index periods. All vouchered specimens should be recorded on the MBSS Specimen Tracking Data Sheet (Appendix G). All preserved and photographed specimens will be reviewed by an expert in taxonomy for each taxonomic group and results will be kept on record. The MBSS Specimen Tracking Data Sheet will serve as a chain-of-custody form between field crew leaders and taxonomic experts.

Taxonomic experts (or a designee assigned by the taxonomic expert) will also audit field identification of organisms. Field audits will be conducted by taxonomic experts (or designee) at a minimum of one site per crew.

3.12 Legibility
To ensure accurate transfer of information from hard copy data sheets to the MBSS data base, data must be recorded on data sheets legibly. If the handwriting of certain individuals is deemed illegible by the crew leader, then those individuals should not record data on data sheets.

4.0 Preparation for Sampling
The purpose of this chapter is to outline procedures and provide guidance for pre-deployment activities to be completed prior to each field sampling trip.

4.1 Equipment
Prior to each field sampling trip, the Crew Leader should ensure that all necessary sampling equipment is prepared for sampling. Equipment lists for sampling during the Spring and Summer Index Periods are provided in the back on this manual.

The Crew Leader will be responsible for ensuring that all necessary equipment and supplies are loaded into the vehicle. The crew will depart for sampling only after the Crew Leader has verified the equipment inventory.

At the end of each sampling day, the Crew Leader will ensure that all sampling equipment is properly stored and that gear, data sheets, preservatives, sample bottles, etc., needed for the next day are identified.

To provide access to unimproved roads and thereby reduce travel time to numerous sample sites, four-wheel drive vehicles should be used when possible for MBSS sampling. Prior to use each day, the Crew Leader will visually inspect the sampling vehicle for any evidence of safety or mechanical problems.

5.0 Sample Collection

5.1 Introduction
The purpose of this chapter is to describe, in detail, the specific procedures that must be followed during sampling for the Round Four MBSS, including water quality, benthic macroinvertebrate, fish, reptile, amphibian, crayfish, mussel, invasive plant, physical habitat, and geomorphology sampling. Sections on site location, sampleability determination, photodocumentation, and temperature logger deployment and retrieval are also included. Strict adherence to all of these protocols is imperative. Of particular importance is diligence in completing and verifying the complete and accurate recording of data sheet information while still in the field and completing sampling during the appropriate Index Period.

5.2 Index Periods

To provide a synoptic view of the current ecological status of Maryland streams, MBSS sampling takes place during two index periods, spring and summer. The Spring Index Period extends from 1 March to 30 April, and the Summer Index Period extends from 1 June to 30 September each year. Four primary activities are conducted during the Spring Index Period: benthic macroinvertebrate, water chemistry for laboratory analysis, select physical habitat variable sampling, and vernal pool searches. During the Summer Index Period, eight primary activities are conducted: fish, reptile and amphibian, stream salamander, mussel, crayfish, invasive plant, select physical habitat, and geomorphology sampling. It is imperative that sampling for these variables be performed during the appropriate index period. Although focused sampling for reptiles and amphibians, crayfishes, and mussels are conducted during the summer index period, incidental observations of any of these taxa should be recorded during any visit to the site, during any time of the year. If no specific place for recording the incidental observation of a particular species is available on data sheets, it should be recorded in the comments section of an available data sheet.

The time period for the Spring Index Period is based on sample degree-day accumulations of mean air temperatures above 4.5°C. This time period was chosen because studies in Maryland have demonstrated that sampling in spring can estimate the degree of acidification in a stream, within acceptable limits, and also provide benthic macroinvertebrate data most suited for identifying anthropogenic stressors at a site.

Based on the results of benthic macroinvertebrate studies, degree day accumulations above certain thresholds (440°C for Coastal Plain and 1050°C for the rest of Maryland) were used as a basis for determining when MBSS Spring Index Period sampling should be completed. Since degree day accumulations rarely approached these thresholds during March and April, the Spring Index Period for Round Four includes all days within these two months. Degree days do not need to be taken into consideration.

The MBSS Summer Index Period was selected to occur during the low flow period, which is most limiting to fishes. Sampling during this period is also advantageous because spawning effects are minimized, temperatures are conducive to wading and water contact, and capture efficiency using electrofishing is typically best when streams are relatively low and warm. The other taxa which MBSS summer sampling documents are most active and/or most easily observed/captured (crayfishes, mussels, stream salamanders, invasive plants) during this time period.

Since water levels are typically at their lowest in Maryland streams during the summer, the Summer Index Period is also the time during which physical habitat is most limiting to many stream dwelling organisms (including fishes, mussels, stream salamanders, and crayfishes). Physical habitat quality and quantity measurements are taken during this time; therefore, they represent limiting conditions for these organisms.

5.3 Site Location

Sites sampled during Round Four will consist of sites that were sampled previously during Round One and Round Two, as well as sites in specific areas that are being used to answer important resource management questions (targeted sites). Sites previously sampled during Round Two are on non-tidal 1st – 4th order (Strahler) streams based on a 1:100,000 scale stream reach file. Sites previously sampled during Round One are on 1st – 3rd order (Strahler) streams based on a 1:250,000 scale stream reach file. Sites from Round One or Two to be re-sampled are randomly chosen from the complete list of sites sampled previously. The complete list of sites sampled previously will be provided to the crew leader with a priority number assigned to each site by watershed. Crew leaders should sample the lowest number (corresponding to the highest priority) sites on the list with permission to sample, by watershed, until the target number of sites in each watershed has been sampled.

All sites consist of the watered portion of the stream and an area 50 meters perpendicular (on both sides) to the stream. Each site is 75 m in length. It is important to sample Round One and Two sites as closely as possible

to locations previously sampled. Crew leaders should use geographic coordinates and previous site location descriptions (as recorded on datasheets in previous Rounds) to re-locate sites. In Rounds One and Two, the 0, 25, 50, and 75 meter transects of these sites were marked with flagging tape and the approximate locations of the 0 and 75 m transects were marked with orange paint on the nearest tree. Note that the flagging tape more precisely marked the transect locations. If markings from Round One or Two are found while accessing the site, the site should be sampled based on those markings. Any additional flagging needed to mark transects should be added and flagging in poor condition should be replaced. If, based on reasonable effort, the markings are not found, then the site location should be determined based on geographic coordinates and, when available, site location descriptions recorded when the site was originally sampled. The coordinates represent the mid-point of the 75 m long site (37.5 m from the downstream end of the site). If arrival at these coordinates occurs and the location is not on a stream, the mid-point of the site should be designated as the point that is reached using the shortest distance to the stream from the location indicated by the GPS. No matter how the site location for Round Four sampling was determined, new geographical coordinates from the mid-point of the site should be recorded on the Spring Habitat Data Sheet; even if they are identical to the coordinates used to find the site. A copy of the MBSS reach file should be consulted following the identification of the site location to be sure that (based on the reach file) the correct stream is being sampled. Maps showing landowner properties (usually tax maps) should also be consulted to ensure that the site is located on a property or properties where landowner permission has been acquired. Permission to use any landowner's property for access to or sampling of any MBSS site is required. In extreme cases, where landowner permission or other sampleability issues prohibit sampling a site in the exact location where the site was chosen, the site may be moved up or downstream no more than one site length distance (75 m) from the original location and substantial documentation must be provided in the comments section of the Spring Index Period Data sheet to justify the location change. This option should be used only after all other options have been exhausted by the Crew Leader. A map showing the location of the site must also be included with data sheets as well as proper landowner permission information.

5.4 Site Selection and Determination of Sampleability

To ensure that a site can be safely and effectively sampled, the Crew Leader will examine the stream prior to the initiation of any sampling. General criteria for determining sampleability include: safety, landowner permission, ability to electrofish effectively, and non-tidal status. No sampling should take place under dangerous conditions. If the site has non-wadeable areas that can be safely sampled using a combination of long-handled anodes and/or dipnets, the site should be considered sampleable. Examples of conditions which could deem a site unsampleable include: a dry stream bed, obvious tidal influence, and unsafe velocities/depths. The determination of sampleability for benthic macroinvertebrates, spring physical habitat assessment, vernal pools, and spring water chemistry should be noted on the Spring Index Period Data Sheet. Sampleability for electrofishing, summer physical habitat assessment, herpetofauna, mussels, and crayfishes should be noted on the Summer Index Period Data Sheet. A description of how to determine sampleability for each of these is included with the description of sampling methods for each. A list of codes for sampleability is provided in Appendix D. If a Round One or Two repeat site is deemed unsampleable during the spring visit, a Spring Index Period Data Sheet should be filled out indicating the reason the site was not sampleable. The next higher priority number site, with permission to sample, should then be sampled in place of the unsampleable site. If a site is unsampleable during the summer visit, another site should not be sampled.

1. Culverts. It should be noted that some sites may still be sampleable even though they include underpasses, beaver dams, large culverts, and dry sections. In the case of small culverts which can not be electrofished, the length of the culvert should be measured and recorded on the data sheet and the length added to the original 75 m site. If the culvert occurs in the first half of the site, the additional distance should be added to the downstream end of the site. Similarly, the additional distance should be added to the upstream end, if the culvert is within the upper half of the original site. If the culvert can be sampled completely, no change should be made to the original 75 m site.

2. Moving Sites. The location of a site (even a randomly selected site) can be changed to ensure that a sample is collected as close as possible to the location originally chosen for sampling. However, the maximum distance that a site should be moved is 75 meters. It is imperative that a randomly selected site be moved as little as possible. Sites (especially randomly selected sites) should only be moved after every attempt has been made to sample the site in its originally chosen location.

5.5 Marking Sites

The 75 m that make up an MBSS site are measured beginning with the 0 m mark at the downstream end of the site and ending with the 75 m mark at the upstream end. At a minimum, the extent of the 75 m site (0 m and 75 m locations) should be clearly marked while sampling is being conducted. During MBSS Rounds One and Two, orange spray paint and flagging were used to mark these locations and flagging was used to mark the 25 m and 50 m locations. The 25 m and 50 m locations need to be identified along with the 75 m and 0 m locations to complete Summer Index Period habitat sampling. Marking of all four locations (0 m, 25 m, 50 m, and 75 m) during the spring index period is recommended. In some rare cases, marking with conspicuous markings, like orange spray paint, may not be allowed or appreciated by landowners. All effort should be made to adequately mark the site in the spring so it can be found again during the summer. To the full extent possible, all flagging or other material used for marking sites should be removed from the site following the last visit to the site. If necessitated by landowner concerns, the orange mark can also be painted over in brown or grey during the summer visit.

5.6 Photographic Documentation

All MBSS sites require at least one photograph be taken of the stream being sampled. Typically, at least two photographs are taken from the mid-point of the site, one looking upstream and one looking downstream. These photographs are typically taken during the Spring Index Period and are used to depict the general appearance and conditions of the stream. Any unusual or unique conditions that exist at the site should be documented with a photograph. Examples of unusual or unique conditions include severely eroded stream banks or trash dumping, pipes or other point source discharges, unusual water coloration, abundant flocculent, large silt or sediment deposition, and riparian tree cutting. Many conditions may warrant taking a photograph to document observations. Crew Leader judgment should be used when deciding what conditions should be photographed. However, when in doubt, take a picture.

A unique number should be used to label each digital photograph on the camera. This number, along with a descriptive title, should be entered in the appropriate portion of the Spring or Summer Index Period Data Sheet, depending on when the photograph(s) was taken. Digital photograph files should be stored with file names that include (at a minimum) the site identification and the unique photograph number. All files should be appropriately backed up.

5.7 Water Chemistry for Laboratory Analysis

Selected water quality variables (pH, acid neutralizing capacity, sulfate, nitrite, nitrate, ammonia, total nitrogen (dissolved and particulate), ortho-phosphate, total phosphorous [dissolved and particulate], chloride, conductivity, dissolved organic carbon, copper, zinc, calcium, and magnesium) are measured based on grab samples taken during the Spring Index Period (1 March to 30 April). These analytes provide information about the state of acidification, degree of organic loading, and specific ions known to influence stream biota. Approximately 1.5 L of water and at least 50 mL in a closed syringe are needed to provide data for all of the MBSS laboratory water chemistry parameters.

The basic protocols used to collect samples in spring follow those used in other DNR-sponsored acid deposition studies, including the Western Maryland Stream Survey and the Western Maryland Watershed Mitigation Study. All bottles for water sampling should have been leached in deionized water for at least 24 hours prior to field

use, and syringes should be new and unopened. All sampling equipment should be carefully packed to eliminate potential contamination. If any contamination is suspected, spare sample bottles or syringes should be used. Water samples for MBSS laboratory analyses should be collected without regard to stream stage and the amount of precipitation or the time since the last precipitation. The only criterion that must be met is that a water sample can be collected safely. However, sampling during turbid conditions or just after heavy rains should be avoided to ensure that benthic habitat can be properly evaluated.

Water must be collected prior to, or upstream of, any disturbance to the stream caused by site sampling or access. Stepping in the stream upstream of the location where water is being collected should be avoided until after all of the water has been collected. Collecting water at the upstream end (75 m) of the site can ensure that other sampling can occur coincident with the collection of water samples. When possible, the area from which water is taken should be near the center of the stream channel, in flowing water, and where adequate depth is present to completely submerge the water sampling bottles.

Each 1 L and 0.5 L sample bottle and syringe must be labeled. The label should include: "MBSS", the date, and site identification as recorded on the top of the Spring Index Period Data Sheet. Each syringe and sample bottle label must be verified by a member of the field crew for accuracy, with verification indicated on the Spring Index Period data sheet. All labels on samples for laboratory analysis should be covered with clear plastic tape to ensure the labels are not smudged or lost. Labels for QC samples below should use letter characters in place of numbers in the segment portion of the label (e.g. 1=A; 2=B, 3=C, etc., and 0=J).

Using care to avoid potential sample contamination from handling, fill the pre-leached 0.5 and 1 liter sample bottles to half-full, rinse, and discard. Repeat the process twice (so that the bottle has been filled and rinsed a total of three times). Then fill the sample containers such that no or a minimum of air space exists in the neck of the bottle. Check to ensure that the seals on both sample bottles are tight.

Place a Luer Lock valve on the end of the syringe. Fill the syringe three times, expelling the water each time. Fill the syringe a fourth time to approximately the 60 ml mark. Hold the syringe in a vertical position and gently tap it until all bubbles are released. After all air is expelled from the syringe, use the plunger to release 5 to 10 ml of sample. When the volume in the syringe is 50 to 55 ml, and while still discharging water, carefully close the Luer Lock valve. Syringes should not contain more than 55 ml of sample to minimize the possibility of plunger dislodgement during shipping or less than 50 ml to provide sufficient water to determine the pH effectively.

Place samples on ice inside a closed lid cooler to maintain samples at 4°C until laboratory analysis is performed.

If a blank sample is to be taken at the site being sampled, that sample should be taken before collecting a routine sample at the randomly selected stream reach. Blanks should be collected following collection procedures outlined above, except that water from the deionized water container should be substituted for stream water. The letter B indicating blank should be entered on the QC label portion of the data sheet. The label for the QC, blank sample should be the same as the original sample, Except that letters should be substituted for numbers in the segment portion of the label (e.g. 1=A; 2=B, 3=C, etc., and 0=J).

If a duplicate sample is to be taken, that duplicate sample should be collected immediately after the routine sample using the same methods described for stream sampling above. The letter D indicating duplicate should be entered on the QC label portion of the data sheet. As with the blank sample, the label for the QC, blank sample should be the same as the original sample, except that letters should be substituted for numbers in the segment portion of the label (e.g. 1=A; 2=B, 3=C, etc., and 0=J).

After sample collections are completed, the field data and chain-of-custody forms (see Appendix G) should be completed and checked by the field crew for completeness and accuracy.

Sample bottles must be shipped to the analytical laboratory via overnight mail within 48 hours of collection.

Special attention should be given to packing samples in such a way that they are unlikely to leak or break during transport. During the packing process, re-verify that data sheets, labels on samples, and chain-of-custody sheets are consistent, and that a complete sample has been taken.

5.8 Physical Habitat

Physical habitat assessments conducted by MBSS are intended to represent the habitat conditions available to the organisms living in the streams and to report on the extent to which certain anthropogenic factors may be affecting Maryland's streams. MBSS habitat assessment protocols are based on a combination of metrics modified and adapted from USEPA's Rapid Bioassessment Protocols (RBP) and Ohio EPA's Qualitative Habitat Evaluation Index (QHEI). Although EPA's RBP habitat assessment protocols differentiate between riffle-run and pool-glide stream types, all metrics selected for the MBSS are scored at all MBSS sample sites to allow direct comparisons across physiographic regions and summaries of conditions on a statewide basis.

Certain MBSS physical habitat variables are recorded based on counts, measurements, or estimates made in the field. These variables include distance from nearest road to site, width of riparian buffer, stream gradient, width, depth, velocity, culvert width and length, extent and height of eroded bank, numbers of woody debris and root wads, extent of channelization, percent embeddedness, and percent shading. The quality of five habitat assessment metric variables along with the severity of bank erosion, buffer breaks, and bar formation are rated using standardized MBSS rating methods. The collection of data on certain other habitat variables are based on the observation (or not) of certain conditions such as buffer breaks, land use types, and evidence of channelization. Based on observations at sites, the absence, presence or extensive presence of stream character and bar substrate is recorded. The type and relative size of riparian vegetation and the type of land cover adjacent to the buffer are reported using standard MBSS codes. The method used for collecting data in the field for each variable differs based on the expected use of each variable as well as optimizing the time required to collect useable information.

Data sheet entries for all physical habitat variables are based on observations within or from the 75 m site only, unless otherwise stated below.

For MBSS physical habitat assessment variables, in all cases where it is necessary to differentiate the left bank of the stream from the right bank, the left and right are determined while facing **upstream.**

Only persons who have attended MBSS training and have demonstrated proficiency with performing MBSS physical habitat assessments should conduct MBSS physical habitat assessments.

Most MBSS physical habitat assessment information is collected during the Summer Index Period. However, a number of important measures are rated during the Spring Index Period. Detailed descriptions of how data are to be recorded for each variable follow. The physical habitat data collected during the Spring Index Period are described first followed by those collected during the Summer Index Period.

Round Four sites that were previously sampled during **Round One** will have four extra parameters assessed - bank stability, minimum buffer width, channel flow status, and remoteness –using the same methods used during Round One. These can be assessed during the Summer Index period. Procedures for their assessment are described in section 3.5.9.2 below (numbers 13-16). Habitat data for Round One repeat sites should be entered on the Round One Repeat Habitat Data Sheet (Appendix H).

5.8.1 Spring Index Period Physical Habitat Assessment

The physical habitat assessment variables recorded during the Spring Index Period can be found on the MBSS

Spring Habitat Data Sheet and should be recorded on this sheet. The methods used to determine exactly what should be recorded for each variable are described, by variable, below. Data sheet entries for all Spring Index Period physical habitat variables are based on observations within or from the 75 m site only.

If the stream cannot be sampled for spring physical habitat assessment, this should be noted on the Spring Index Period Data Sheet. Codes designating reasons that a stream could not be sampled are provided in Appendix D.

1. **Trash Rating.** The trash rating is scored on a 0-20 scale based on criteria found on the Stream Habitat Assessment Guidance Sheet (Appendix E).

2. **Distance of Nearest Road to Site.** This variable should be measured when practical with a tape measure or GPS to the nearest meter. If it is not practical to measure this distance, it can be estimated to the nearest 10 m.

3. **Riparian Buffer Width.** The riparian buffer width should be measured to the nearest meter on each side of the stream, beginning at the water's edge. The left and right banks of the stream are determined while facing upstream. The average width of the buffer should be recorded. Buffer breaks should not be considered when estimating the average buffer width as buffer breaks are recorded in a different portion of the data sheet (see number 6 below). If the average buffer width is greater than or equal to 50 meters, enter 50 for the buffer width.

4. **Adjacent Land Cover.** Using the codes for adjacent land cover types (found in the back of this manual), the type of land cover immediately adjacent to the stream buffer should be recorded. If the buffer is 50 m or more, then the same code that was recorded for the buffer should be recorded for the adjacent land cover.

5. **Riparian Vegetation.** Using the codes for vegetation types (Appendix D) the dominant vegetation types present within the 50m buffer of the 75 m site should be recorded. As many as four types can be recorded. The vegetation types are recorded in order of their dominance within the buffer, with the most dominant recorded first (in the left most box under the bank where the buffer is being recorded). Stem density and canopy density should both be taken into consideration for determining density. However, stem density should take precedence over canopy density.

6. **Buffer Breaks.** Both banks of the stream for the entire 75 m site should be examined for buffer breaks. For each bank of the stream, if any buffer breaks are observed, then a "Y" should be placed in the box on the MBSS Spring Habitat Data Sheet next to the words "Buffer Breaks (Y/N)". If no buffer breaks are observed, write an "N" in the box.

7. **Buffer Break Types.** If a buffer break is observed while examining the stream banks, the severity of the buffer break should be noted and recorded as M (minor) or S (severe) in the box alongside the most appropriate buffer break type listed on the Spring Habitat Data Sheet.

8. **Channelization.** The site should be inspected for any evidence of channel straightening or dredging. If evidence of channel straightening or dredging are observed anywhere within the 75 m site, the linear extent of the channelization should be measured to the nearest meter. Channelization along each bank and the stream bottom should be measured separately and recorded in the appropriate portion of the Spring Habitat Data Sheet, where the type of channelization is listed. If channelization is observed at a site with a braided stream channel, the total extent of stream channel that is channelized should be recorded. It is possible (when multiple channels are present), using this method, for the total extent of left bank, right bank, or stream bottom channelized to be more than 75 m. Since the objective of this measure is to determine the total length of stream channel that is channelized, this is acceptable.

9. **Land Use.** While at the site, a survey of the surrounding area for land use types is conducted. For each land use type listed on the Spring Habitat Data Sheet mark a "Y" or "N" indicating whether or not the land use type is present near the site. Any land use that can be observed while in or alongside the stream at the site should receive a "Y" and any that cannot be observed should receive an "N".

10. **Stream Gradient.** The intent of this is to measure the slope of the stream over 75 m. This is achieved by recording the difference in water surface height from the 0 m to the 75 m locations of the MBSS site as compared to a level plane. A levelometer was used during the Round Two MBSS to measure stream gradient. Laser levels and other techniques may provide similar results, sometimes with increased precision. Any technique used to measure stream gradient should provide data accurate to at least the nearest 5 centimeters to be comparable to data collected since Round Two at MBSS sites.

    The calibration and proper functioning of the instrument that is used for determining stream gradient must be verified at least once every week while sampling is being conducted and documentation showing verification must be kept with the instrument.

    Measurements of height should be taken from the water's surface and NOT from the stream bottom or any bank locations. Measurements can be taken at a number of locations if the 0 m and 75 m locations cannot be seen at the same time, from the same location. However, if the level must be relocated, height measurements must be taken again from the next closest location where a measurement was already taken.

    If a culvert is present within the MBSS site and the stream level drops below the culvert due to the presence of the culvert, then the stream gradient should be measured without considering the unnatural drop caused by the culvert. This requires two separate sets of height measurements, one downstream from the culvert and one upstream of the culvert. The height difference over the span of the culvert should not be measured in this case.

    Record the height differences that will be used to calculate stream gradient on the Spring Habitat Data Sheet.

11. **Road Culvert.** If a road culvert is present within the 75 m site, an assessment of whether or not the culvert will be sampleable for fish is conducted. The width and length of the culvert should also be measured and recorded on the Spring Habitat Data Sheet.

5.8.2 Summer Index Period Physical Habitat Assessment

The physical habitat assessment variables recorded during the Summer Index Period can be found on the MBSS Summer Habitat Data Sheet and should be recorded on this sheet. The methods used to determine exactly what should be recorded for each variable are described, by variable, below. Data sheet entries for all Summer Index Period physical habitat variables are based on observations within or from the 75 m site only, unless otherwise specified.

In all cases where it is necessary to differentiate the left bank of the stream from the right bank, the left and right are determined while facing upstream.

Many of the summer physical habitat assessment measures require sufficiently clear water to observe the stream bottom throughout the majority of the 75 m site. If conditions do not allow sufficient visibility to see all of the features that must be observed, or if conditions are unsafe for wading, the site should be considered unsampleable for physical habitat. In many cases, the stream may be sampleable during a return visit when the water level is lower. However, if the stream cannot be sampled for summer physical habitat assessment, this should be noted

on the Summer Index Period Data Sheet. Codes designating reasons that a stream could not be sampled are provided in the back of this manual.

1. Habitat Assessment Metrics. Five metrics: instream habitat, epifaunal substrate, pool quality, riffle quality, and velocity depth diversity are rated on a scale of 0-20 using criteria provided on the Habitat Assessment Guidance Sheet (in the back of this manual). The scores for each of these metrics are meant to characterize a distinct aspect of stream habitat. The instream habitat metric primarily addresses habitat for fishes and epifaunal substrate is meant to rate the suitability of habitat for benthic macroinvertebrates. The general quality of riffle and pool habitats are rated based primarily on the prevalence of sufficient depth and extent of these habitats. Velocity/depth/diversity provides a measure of the how well fast, slow, deep, and shallow areas are represented in the stream.

2. **Embeddedness.** The percent of coarse riffle substrates surrounded by fine substrates, such as sand and silt, is recorded based on visual observation. Riffle substrates that are examined should include the area with the fastest flow within riffle or run habitats. If no riffle is present within the 75 m site, embeddedness can be rated based on the closest available riffle located in the same reach as the site (but should not be more than 75 m away from the upstream or downstream end of the site). Several substrates should be examined within the riffle to determine the approximate average condition within the fast part of the riffle. Substrates should be examined for embeddedness prior to disturbances (such as walking or netting) that are likely to dislodge fine materials from around larger substrate.

3. **Shading.** The percent of the wetted area of the 75 m site that is shaded by overhanging vegetation or other structures is approximated based on a visual assessment. If clearing of vegetation was conducted to facilitate electrofishing, or for any other reason, shading should be rated based on the condition prior to clearing.

4. **Woody Debris.** For the MBSS, large woody debris are defined as any natural woody structures (e.g. logs, snags, dead tree trunks), with the exception of live trees that are at least 10 cm in diameter and more than 1.5 m long. The number of large woody debris, located in the wetted portion of the 75 m stream site (instream woody debris), is counted. The number of large woody debris in the stream channel or immediate riparian area, but not in the wetted portion of the stream (dewatered woody debris) are counted separately from instream woody debris. Only those dewatered woody debris from the immediate riparian area that (in the opinion of the evaluator) are likely to become wetted during high flows, or fall into the stream channel should be counted.

5. **Root Wads.** For the MBSS, root wads that are on live trees with a chest high trunk diameter (DBH) of at least 16 cm should be counted. These should be counted along both banks of the stream within the 75 m site. Those root wads that are in the water (instream) are counted separately from those not in the stream (dewatered). However, only those dewatered root wads that provide stability to the stream bank or that are likely to become wetted during high flows should be counted.

6. **Stream Character.** The Stream Character portion of the MBSS Summer Habitat Data Sheet lists 15 stream features. For each feature, an A, P, or E should be recorded in the box next to the feature indicating whether the feature is absent, present, or extensive respectively in the 75 m stream site.

7. **Maximum Depth.** The maximum depth of the MBSS site is considered the deepest area found anywhere within the 75 m. Maximum depth is recorded to the nearest cm.

8. **Wetted Width, Thalweg Depth, and Thalweg Velocity.** The wetted width, thalweg depth and thalweg velocity are measured at four transects within the 75 m MBSS site. The four transects are located at the 0 m, 25 m, 50 m, and 75 m portions of the MBSS site (beginning with 0m at the downstream-most end of

the site). Wetted width is measured from bank to bank (perpendicular to the direction of the stream flow) to the nearest 0.1 m and includes only the wetted portion of the stream. Islands or other large features in the stream that would not be covered by water during higher base-flow should not be included in the measurement of wetted width. Features that would be covered by water (during higher base-flow should be included in the wetted width measurement. Thalweg depth is the depth (in cm) of the deepest part of the stream at each transect. Thalweg velocity is the stream current velocity (in m/sec) in the deepest part of the stream at each transect.

9. **Flow.** Measurements that can be used to calculate flow (often referred to as discharge) are recorded on the MBSS Summer Habitat Data Sheet. A transect that is suitable for taking these measurements should be located. A suitable transect approximates a "U" shaped channel to the extent possible. The most useful measurements are acquired by avoiding transects with boulders or other irregularities that create backflows and cross flows. The stream channel can be modified to more closely approximate a "U" shaped channel and provide laminar flow with adequate depth for taking velocity measurements. Unless the stream is very small (less than 0.5 m wide), a minimum of 10 measurements should be taken. As many as 25 measurements can be recorded on the MBSS Summer Habitat Data Sheet. In general, more measurements are required in larger streams. The measurements consist of depth (to the nearest 0.5 cm) and velocity (to the nearest 0.001 m/sec) and should be recorded at regular intervals. Velocity measurements should be taken at 0.6 of the distance from the water surface to the bottom (measured from the surface), making sure to orient the sensor to face upstream and taking care to stand well downstream to avoid deflection of flows. Depth and velocity measurements should be taken at the exact same locations. The "Lat Loc" on the MBSS Summer Habitat Data Sheet refers to the distance from one stream bank (either left or right) where each depth and velocity measurement is taken.

10. **Alternative Flow.** If flows are so low that they can not be measured with a flow meter, the stream should be constricted as much as possible in a 1 meter section of uniform width and depth. The speed of a floated object should be recorded three times as a substitute for velocity measured with the flow meter. Record on the data sheet the depth, width, and time (3 trials) for the floated object.

11. **Bank Erosion.** The length and average height of erosion on both banks of the stream, within the 75 m site should be recorded along with the severity of erosion, on the MBSS Summer Habitat Data Sheet. In braided streams it is possible to have the total extent of eroded bank add up to more than 75 m. Since the objective of this measure is to determine the total area of erosion present at the site, this is acceptable.

12. **Bar Formation and Substrate.** Boxes in this portion of the MBSS Summer Habitat Data Sheet should be filled in completely to indicate if the bar formation is absent (fill in the box next to "None"), minor, moderate, or extensive; and the dominant particle type(s) that make up the bars in the site. More than one particle type can be selected. However particles comprising only a minor part of the substrate should not be selected. Dominance of particle types should be determined based on the proportion of the bar consisting of each type.

13. **Round One Repeat of Bank Stability.** Bank stability as it was assessed during Round One consists of scoring on a 0-20 scale, as with many of the other MBSS physical habitat assessment metrics. Guidance for scoring comes from the MBSS Round One sampling Manual as follows: A score of 16 – 20 if the upper bank is stable, 0-10% of banks with erosional scars and little potential for future problems; A score of 11 – 15 if banks are moderately stable, 10 – 30% of banks with erosional scars, mostly healed over, with slight potential for problems in extreme floods; A score of 6 – 10 if banks are moderately unstable, 30-60% of banks with erosional scars and high erosion potential during extreme high flows; A score of 0 – 5 if the banks are unstable, raw areas frequent along straight sections and bends and side slopes > than 60o common.

14. **Round One Repeat of Channel Flow Status.** Channel Alteration as it was assessed during Round One consists of scoring on a 0-20 scale, as with many of the other MBSS physical habitat assessment metrics. Guidance for scoring comes from the MBSS Round One sampling Manual as follows: A score of 16 – 20 if there is little to no enlargement of islands or point bars, no evidence of channel straightening or dredging, 0-10% of stream banks artificially armored or lined; A score of 11 - 15 if bar formation is mostly from coarse gravel and/or 10-40% of stream banks are artificially armored or obviously channelized; A score of 6 – 10 if recent but moderate deposition of gravel and coarse sand is on bars and/or embankments on both banks, and/or 40-80% of banks are artificially armored (or channel lined in concrete); A score of 0 – 5 if there is heavy deposition of fine material, extensive bar development, OR recent channelization or dredging evident, or over 80% of the banks are artificially armored.

15. **Round One Repeat Remoteness.** Round One Repeat of Remoteness as it was assessed during Round One consists of scoring on a 0-20 scale, as with many of the other MBSS physical habitat assessment metrics. Guidance for scoring comes from the MBSS Round One sampling Manual as follows: A score of 16 – 20 if the site is more than ¼ mile from the nearest road, access difficult and little or no evidence of human activity; A score of 11- 15 if the site is with ¼ mile of (but not immediately accessible to) the road and is accessed by trail and the site has moderately "wild" character; A score of 6 – 10 if the site is within ¼ mile of road and accessible by trail and human activities are readily evident; a score of 0 – 5 if the site is immediately adjacent to a road with obvious human activities evident.

16. **Round One Repeat of Minimum Buffer Width.** The minimum buffer width for repeated Round One sites should be assessed the same way as number 3 under section 3.5.9.1 above, except that the minimum width of the buffer for each bank should be recorded. Rather than recording "buffer breaks", a break in the buffer indicates no buffer at all. Adjacent land cover types and buffer vegetation should be recorded using the same codes as referenced in section 3.5.9.1 number 3.

5.9 Geomorphology Sampling

5.9.1 Recommended Equipment – MBSS Geomorphology Sampling

Cross-section datasheets printed on Rite-in-the-Rain paper
Riffle pebble count datasheet printed on Rite-in-the-Rain paper
Regional curve derived bankfull parameters
Clipboard
Pencil
Tripod
Self-leveling laser level and audible laser receiver
Top-setting, telescoping survey rod marked with hundredths of a foot
300-foot measuring tape marked with tenths of a foot
Silvey Stakes
Pin flags
Metric ruler/meter stick (with mm markings) (1 per surveyor) or Gravelometer (1 per surveyor)
Sand gauge reference cards (1 per surveyor)
Metal hand tally counter (clicker) (1 per surveyor)
Digital camera
Hand shears, machete or other clearing device

5.9.2 Physical Stream Channel Measurement

The goal of physical stream channel measurement at MBSS sites is to collect channel dimension measurements and particle size in a representative riffle or straight reach at each site. The measurements will be taken using

standard surveying and pebble count techniques. Cross-section and pebble count data will be collected during the Summer Index Period.

5.9.2.1 Cross-Section Survey Protocol

1. **Survey Site Selection.** Within the site, choose a representative riffle area to set up the cross-section. The area should be free from direct anthropogenic alterations and reflective of local geology such that the stream is able to adjust its banks under its current flow regime. The riffle location should be chosen along a relatively straight stretch of the stream when possible. Avoid transverse riffles, riffles located at bends, riffles directly influenced by tributaries/confluences, etc. Locate the cross-section within the top-third of the riffle, below the riffle crest (peak). The riffle crest is defined as the highest elevation within that specific riffle. Establish the cross-section perpendicular to the direction of flow. If no riffle is present within the site (e.g., some Coastal Plain streams), choose a cross-section within a relatively straight portion of the stream that is most representative of the site conditions and where flow is present. Mark on the Cross-Section Data Sheet whether or not the cross-section is located in a riffle. If no riffle is present in the site but present outside of the site, it is acceptable to use the out-of-site riffle as long as the crew has permission along both banks and there are no tributaries between the MBSS site and the out-of-site riffle. Mark on the Cross-Section Data Sheet if the selected riffle is located outside of the 75m MBSS site by placing a "Y" in the appropriate box.

2. **Cross-Section Surveying.** Stretch the tape (marked in tenths of feet) across the channel (zero on right bank facing upstream) making sure the tape is perpendicular to the direction of flow. The tape should be taut and should extend away from the stream for one channel width from the top of each bank. Use Silvey stakes or something similar to secure each end of the tape. Channel width will be measured from top of the right bank to top of the left bank. The minimum width of the cross-section will be three channel widths. In many cases, this will cover the flood-prone area (area bordering the stream that will be covered by stream waters at a flood stage of twice the thalweg bankfull depth). The flood-prone area width will be measured after the cross-section has been surveyed following the directions given in 5.9.1.2.3 below.

   Setup the surveying instrument in a location where the entire cross-section can be viewed. Vegetation may be pruned to allow the entire cross-section to be surveyed without moving the laser and tripod. The instrument should be placed at an elevation higher than the highest feature (e.g., flood-prone elevation) required for the survey. Ideally, only one instrument setup will be required to survey the entire cross-section; however, determining the width of the flood-prone area may require multiple instrument setups if foliage is dense.

   Use the pre-determined mean bankfull depth value obtained from a regional curve within the riffle cross-section to identify the bankfull stage on left and right banks. Additional field determinations of bankfull may be recorded if the field crew observes obvious bankfull features. Field bankfull determination is optional.

   Survey the elevation of station zero on the right bank looking upstream. Station 0 will be approximately one channel width upland from the top of the right bank. Place the rod firmly on top of the ground and hold it as steady and vertical/plumb as possible while moving the receiver up/down until the audible tone indicates a proper reading. Once the elevation is determined, the person holding the rod should call out to the person recording data the reading from the rod in hundredths of feet. Record this as station zero and the corresponding elevation in hundredths of feet on the datasheet.

   Continue surveying across the cross-section obtaining rod readings at major breaks in bed elevation (Figure 1). These readings will all be recorded on the Cross-Section Data Sheet. Typically, 15 to 20 points are necessary including key features such as top of right bank (TOB-R), right bankfull (RBF), right edge water (REW), Thalweg (THL), left edge water (LEW), left bankfull (LBF) and top of left bank (TOB-L). Other significant depositional features or breaks in slope should also be surveyed. Record the distance on the tape

(station), the corresponding rod height and feature notes on the cross-section datasheet. Record station measurements in tenths of feet and rod heights in hundredths of feet.

The final measurement of the cross-section survey will be a second elevation at station zero. This second measurement of station zero should be used as a QA/QC check of the survey data.

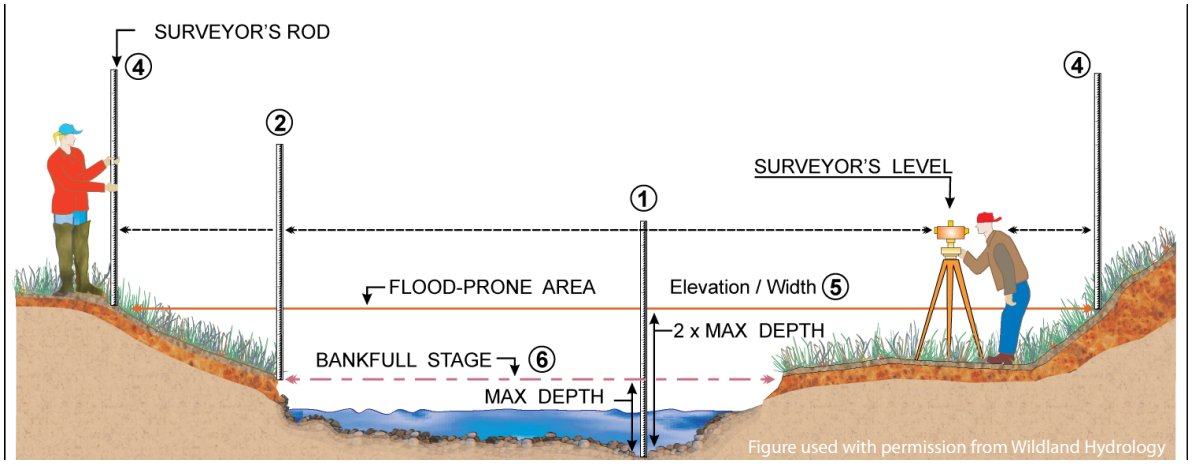

Figure 1 - Surveying a Cross-section.

If banks are severely undercut or slumping, an additional measuring device (e.g., measuring rod or yard stick equipped with a small level) can be used as a base for the main surveying rod. One person should hold the second rod horizontally against the bank at the first location of undercut, and perpendicular to the main survey rod, keeping it as level and steady as possible. A second person should rest the main survey rod on top of the second rod, making note of the distance of the base of the main rod along the second rod (reading 1) and the stationing of the main rod on the survey tape where they cross (reading 2). Move the receiver on the main rod up/down until the audible tone indicates a proper reading and record the elevation. Then determine stationing for the undercut by subtracting reading 1 from reading 2. Then move the entire setup down to the next point of measurement on the undercut bank and repeat until the bank is no longer undercut.

3. **Determining Flood-Prone Area.** Find the station on the cross-section that has an average water depth for that cross-section and take an elevation of the streambed. The elevation at that station will be entered on the Flood-prone Area Data Sheet. The mean bankfull depth value from the regional curve will be entered next on the Flood-prone Area Data Sheet and subtracted from the elevation at average water depth. This results in the bankfull elevation for this cross-section.

   The thalweg elevation from the cross-section should be transferred from the Cross-Section Data Sheet onto the Flood-prone Area Data Sheet. The bankfull elevation will be entered next and subtracted from the thalweg elevation. This results in the maximum bankfull depth for this cross-section. The maximum bankfull depth should be multiplied by two and entered into the 2x Max Bankfull Depth box.

   The thalweg elevation should be entered again on the Flood-prone Area Data Sheet. The 2x max bankfull value should be transferred into the next box and subtracted from the thalweg depth. This results in the flood-prone area elevation.

   The flood-prone area elevation should be used to determine the width of the flood-prone area of the cross-section. If this elevation is captured on both sides of the stream in the cross-section then the survey is complete. If the cross-section on neither side, or only one side contains the flood-prone elevation, then two additional survey points need to be collected.

The surveying rod should be set to the calculated flood-prone elevation. The person holding the rod should then traverse away from the stream, staying in line with the cross-section, until the preset surveying rod measures the flood-prone area elevation when set on the ground. This location is marked with a pin flag. This process is completed again on the opposite side of the stream. A tape is stretched between the two pin flags and the resulting distance is recorded as the width of the flood-prone area.

If the flood-prone elevation is greater than 50m from the edge of the stream, it is marked as greater than 50m on that side of the stream on the Flood-prone Area Data Sheet and the width is not measured in the field.

4. **Photodocumentation.** Take four photographs of each cross-section and record the photograph number, time stamp, and location information on the datasheet. Take one photograph from upstream of the cross-section looking downstream, one from downstream of the cross-section looking upstream, one from the left bank looking at the right bank, and one from the right bank looking at the left bank.

5. **Field QA/QC.** While the measuring tape is still deployed and level is still set up, the field crew leader must make sure that all measurements have been recorded on a data sheet and photographs taken. Recorded data must be reviewed at the point of entry. The crew leader and one other member of the crew must review and initial all field datasheets. Any errors found should be corrected before leaving the site.

5.9.2.2 Riffle Pebble Count Protocol

1. **Locating the Pebble Count.** The riffle pebble count should be conducted within the riffle or other section of stream channel used for the riffle cross-section survey.

2. **Riffle Pebble Count Protocol.** Ten transects (perpendicular to flow) within the riffle should be sampled for pebbles. Each transect will start at the wetted edge of the riffle on one bank and continue across the wetted portion of the riffle, ending at the edge of the opposite bank. Distribute transects for the riffle pebble counts evenly along the entire riffle feature. Sample each transect moving perpendicular to the stream banks until 10 random particles equally spaced along that transect have been measured. Repeat this procedure until 10 random particles at each of 10 different transects have been measured for a total of 100 particles. A metal hand tally counter/clicker can be used to help keep track of transect counts. To avoid bias of selecting larger particles, the observer should look away from the channel bed and select the first particle touched by the tip of index finger at observer's toe. While traversing across a transect, it is possible to select the same large piece of substrate more than one time. It is acceptable to measure the same large piece of substrate more than once.

3. **Measuring the Particle.** Measure the length of the intermediate axis in millimeters and mark a dot in the correct column and row on the data sheet. (The intermediate axis is neither the longest nor the shortest of the three mutually perpendicular sides of the particle). If the particle is linear-shaped, average the axes. If the particle is very small and a measurement cannot be taken (e.g., sand or silt), sand gauge reference cards can help the surveyor classify the particle appropriately. Do not measure organic material or trash/refuse. If the first particle touched is fine sediment forming a thin veneer on a larger particle, the larger particle is what is measured. The intent is to measure the particles that are defining the roughness of the stream channel.

4. **Field QA/QC.** Tally up counts in each cell of the datasheet to ensure that a total of at least 100 particles were measured and recorded. Recorded data must be reviewed at the point of entry and the crew leader and one other member of the crew must review and initial all field datasheets.

5.9.3 Facies Mapping

The objective of facies mapping at MBSS sites is to characterize substrate type and composition at each site.

Facies mapping data will be collected during the Spring Index Period and recorded on the MBSS Spring Facies Map Data Sheet.

5.9.3.1 Facies Mapping Protocol

1. **Creating the cells.** Divide the MBSS site into 6 cells, based on the 0M, 25M, 50M, and 75M markings, and the center of the wetted channel as follows:

2. **Classifying the substrate.** Record the dominant and subdominant substrate within each cell using the classifications provided on the MBSS Spring Facies Map Data Sheet. The dominant substrate is the category which covers the largest percentage of the cell's area. The subdominant substrate is the category which covers the second largest percentage of the cell's area. A meter stick can be used to measure the intermediate axis of several substrate particles to help classify the size of the substrate.

3. **Classifying stream depth.** Estimate the category of average water depth within each cell using the classifications provided on the MBSS Spring Facies Map Data Sheet. If the average depth is less than 0.5 m

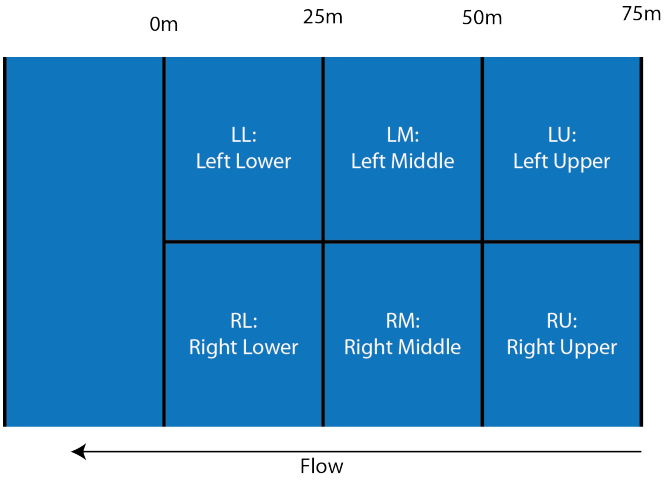

Figure 2 - Left and right are defined while facing upstream.

the category is Shallow; between 0.5 m and 1.0 m the category is Moderately Deep; greater than 1.0 m the category is Deep. A meter stick can be used to take multiple measurements in the cell to obtain an average stream depth for that cell.

4. **Classifying stream water velocity.** Estimate the average stream velocity within each cell. Record the velocity category on the MBSS Spring Facies Map Data Sheet using the categories provided on the data sheet. If the average water velocity is between 0 and 0.3 m/s, it is in the Slow velocity category. If the average water velocity is greater than 0.3 m/s, it is in the Fast velocity category.

5.10 Stream Blockages

Barriers to migration (such as stream blockages) often restrict the movements of resident, as well as diadromous, fishes. The Department of Natural Resources Fisheries Service keeps track of all known barriers to fish migration. The MBSS has provided the locations of many man-made barriers to fish migration to Fisheries Service to aid in documenting their locations so that the most effective possible plans to provide passage can be implemented.

To continue to provide this useful information, any man-made stream blockages either at the MBSS site or en route to the MBSS site, should have the height (to the nearest 0.1 m) and location (latitude and longitude in decimal degrees) recorded on the MBSS Spring Habitat Data Sheet. The type of blockages should also be recorded. Codes for blockage types are provided in the back of the manual. Obvious blockages such as dams on

major rivers need not be recorded, but if there is any doubt about whether or not to record a blockage, recording the blockage is recommended.

5.11 Temperature Loggers

Temperature loggers should be deployed to measure water and air temperature at all MBSS sites. The loggers should be programmed to record temperatures from 1 June to at least 15 August. Each logger should be set to record the highest temperature during an interval not to exceed 20 minutes in duration (shorter durations can be achieved depending on the memory capacity of the logger). Temperature loggers should be deployed within the limits of the sample site, preferably along a bank. The serial number of the temperature logger deployed at each site should be recorded on the MBSS Spring Index Period Data Sheet along with a description of the location where the logger was deployed. Water loggers should be secured to a well anchored tree root, gabion, or other stable structure. Care should be taken when selecting the deployment location to ensure that the temperature logger is not in an area with fast current and that it is placed at a depth to ensure that it will remain submerged until time of retrieval. Temperature loggers deployed to record air temperature should be as close as possible to the location of the temperature logger deployed to record water temperature. The air logger should be at least 3 feet off the ground and no more than 20 meters from the wetted edge of the stream. The air logger can be affixed to a tree or other stable structure. When each temperature logger is retrieved, the time and date of retrieval should be recorded. Verifying that the serial number for the logger that was retrieved matches the serial number entered on the Spring Index Period Data Sheet is recommended. It is often useful (and recommended) to attach a flag or piece of tape to the logger with the site identification, date, and time of retrieval.

5.12 Vernal Pools

1. **Definition.** A vernal pool is a small, temporary body of water that provides vitally important habitat for many amphibians and aquatic invertebrates. Despite their importance, very limited information is currently available on Maryland's vernal pools. Since the beginning of Round Three (2007), the MBSS has added the collection of information on the location and size of potential vernal pool habitats encountered, as well as a list of herpetofauna associated with the pool and minimal physical habitat information. Habitats that qualify as potential vernal pools are less than one acre (4,000 m2), can be very small (less than one square meter), and are not directly connected to a flowing stream.

2. **Index Period.** Vernal pool sampling should take place during the Spring Index Period.

3. **Vernal Pool Searches.** A search for vernal pools should be conducted within the area adjacent to MBSS sites to 50 m perpendicular to each side of the stream and for the entire 75 m length of the site. If an area cannot be searched, the appropriate code is recorded on the data sheet. Examples of conditions that would prohibit or limit searches include areas without permission on one side of the stream and extensive multiflora rose along the stream. If a portion of the area cannot be searched, the approximate unsearchable area should be recorded on the comments section of the data sheet and the appropriate unsampleability code recorded. Vernal pool sampling is only deemed unsampleable if the entire 50 m area on both sides of the stream being searched is unsampleable. If less than the entire 50 m area is sampleable, the approximate area that cannot be searched should be recorded (with a description of the reason it could not be searched) in the comments section of the data sheet. Any vernal pool found in the 50 m area should be recorded on the Vernal Pool Data Sheet and a "Y" should be recorded in the box on the Vernal Pool Data Sheet next to the "Within Transect Y/N" section.

4. **Incidental Findings.** Any vernal pools encountered outside of the transect (the area within 50 m perpendicular to the 75 m site on both sides of the stream) should be recorded on a Vernal Pool Data sheet

and an "N" recorded in the "Within Transect Y/N" section.

5. **Vernal Pool Data.** Geographic coordinates (decimal degrees) should be recorded on the MBSS Vernal Pool Data Sheet for pools that are not within the 50 m transect. No coordinates should be recorded for vernal pools within the 50 m area searched for pools adjacent to the stream site. At least one digital photograph of each vernal pool should be taken and the number of the photograph recorded. If a large number of pools are found in close proximity to one another, one photograph that shows this is sufficient. A large group of pools within sight of one another can also be recorded on one section of the vernal pool data sheet as a vernal pool complex. The approximate dimensions of the potential vernal pool, or pool complex (length, width, and depth) should also be estimated and recorded, along with whether or not the pool is in the floodplain of a stream or not (upland pool). The position of the pools (floodplain or upland is recorded on the data sheet. The land cover adjacent to the potential vernal pool or pool complex should be recorded using the codes provided in the back of the manual. Up to three codes can be recorded. The codes should be entered in order, from left to right on the data sheet, starting with the closest land cover to the pool and ending with the land cover that is furthest away from the pool. The land cover types that are recorded should be the dominant types that are in the area that can be seen while standing next to the pool. The presence of fishes or fairy shrimp (order Anostraca) (observed while standing near the pool) should be recorded using a "Y" for presence and an "N" for not observed. It is not necessary to sample for fishes or other aquatic organisms in potential vernal pools. Any amphibians observed in or immediately adjacent to the potential vernal pool should also be recorded along with the life stage of the organism (egg, larval, juvenile, adult). Frogs that are heard calling from within or immediately adjacent to the potential vernal pool, but that are not observed should also be recorded, with the appropriate box indicating that the species was heard, but not seen, checked next to the species name. Only persons who have passed the MBSS amphibian taxonomic identification test should identify amphibians that are observed and only persons who have passed the frog call test should identify frogs based on calls that are heard.

6. **Wading in Vernal Pools.** Wading in vernal pools with waders that have been in other water bodies can spread diseases that may be highly deleterious to amphibians that use these habitats. In most cases, collecting all of the information described above can be conducted without wading into the water. Wading in potential vernal pools should be avoided.

5.13 Biological Sampling

Biological sampling has always been the primary focus of the MBSS. During Rounds One and Two, the MBSS focused primarily on fish and benthic macroinvertebrates. Indices of Biological Integrity (IBIs) for these groups were developed using Round One and Round Two MBSS data. These IBIs are now the basis of biocriteria in Maryland and have been extensively used to represent the ecological condition of streams and rivers. A stream salamander IBI was also developed using MBSS sampling data along with supplemental sampling. In addition to providing IBI scores, the MBSS is well known for providing the best possible information on fish, benthic macroinvertebrates and stream salamanders available in Maryland. New distributional records for many species (including rare, threatened, and endangered taxa) have been documented by MBSS. MBSS species specific data have also been used to determine biodiversity priority areas so that effective conservation measures can be implemented. Threats and stressors to biota have also been determined from MBSS data and can be used to implement restoration and protection.

The key to the successes that the MBSS has had with biological data has been the consistency that comes from strict adherence to established sampling protocols and the quality control program which ensures (and

documents) that those personnel collecting biological data in the field and laboratory are proficient with taxonomic identification.

Nationally, freshwater mussels and crayfishes are the most imperiled animal groups. A great deal of information is needed on these two groups in Maryland. To help meet this need, crayfish and mussel information will continue to be collected during Round Four. The sampling of stream salamanders has also been expanded to allow more rigorous examinations of salamander information collected at MBSS sites. The information collected on these taxa will provide a great deal of information that will supplement our knowledge of Maryland's biological integrity and biodiversity and will continue to provide much needed information to plan and implement effective restoration and protection measures for aquatic biota in our state.

This section describes the protocols that will be used during the Round Four MBSS for the collection of biological variables.

5.13.1 Benthic Macroinvertebrates

Benthic macroinvertebrate sampling is conducted within the same 75 m site used for other MBSS sampling. MBSS benthic macroinvertebrate sampling must occur during the Spring Index Period (1 March to 30 April). The intent of benthic sampling is to provide a representative sample of the community composition and relative abundance in favorable habitat (habitats supporting the greatest benthic diversity) within the site. In addition to representing the diversity at a MBSS site, benthic macroinvertebrate data collected by MBSS are used to calculate the MBSS benthic macroinvertebrate IBI. A D-net (540µ mesh; 10 inch "D" height), sieve bucket (540µ mesh sieve), and sample bucket are needed to collect an MBSS benthic macroinvertebrate sample.

1. **Sampleability.** Before sampling benthic macroinvertebrates, the crew leader (with input from other crew members) must determine if the site can be sampled safely and effectively. Sampling can only be conducted safely if the site being sampled is wadeable. If the depth or current velocity precludes safe wading, the site should be considered unsafe and not sampleable.

   Effective MBSS benthic macroinvertebrate sampling requires inspection of suitable habitats. Although turbidity or darkly stained water should not prohibit benthic macroinvertebrate sampling, streams that are so turbid that benthic macroinvertebrate habitat cannot be seen at all should not be sampled. Exceptions are sites with persistent and excessive turbidity problems (based on many return visits none of the stream bottom is ever visible). In these cases a note describing the turbidity problem should be made in the comments section of the spring data sheet. Other situations that may preclude sampling include dry streams, marshes, impoundments such as those produced by beaver dams that are too deep to sample, tidally influenced streams, and areas where landowner permission is denied. The appropriate code for unsampleability should be recorded on the Spring Index Period Data Sheet.

2. **Habitats to be Sampled.** Sampling should be conducted at a combination of habitats that support the most diverse macroinvertebrate community within a site. These habitats often include riffles when one is present. Other habitats, in order of preference, are root wads, root mats and woody debris and associated snag habitat; leaf packs; submerged macrophytes and associated substrate; and undercut banks. Other less preferred habitats include gravel, broken peat, clay lumps and detrital or sand areas in runs. Note that, among all the habitats listed above, those that are most stable and in moving water are preferred to those that are unstable and in still water.

3. **Benthic Macroinvertebrate Sampling Protocols.** Benthic sample buckets must be labeled twice - on the external wall of the bucket and on the inside. The following information must be included on the label: date, time, and site identification code from the Spring Index Period Data Sheet. Verify the information on each label and indicate so on the Spring Index Period Data Sheet. The external label should be covered with clear

plastic tape to prevent smudging and/or label loss. Internal labels must be printed on waterproof paper. Both labels should be filled in with pencil. Benthic sample Chain-of-Custody forms (Appendix G) should also be filled out with the name of the sampler, date, time, and sample site number.

Immediately before sampling for macroinvertebrates at each site, ensure that there are no holes or remnants of prior samples in the D-net. Holes must be repaired before sampling commences.

Survey the site to locate the most productive benthic macroinvertebrate habitats as listed above. Twenty square feet of habitat should be sampled at each site and material collected for this 20 ft2 sample is pooled into one sample bucket. Conveniently, the standard D-net used for MBSS sampling is about one foot wide, allowing for easy approximation of the necessary 20 ft2 sample.

The most productive habitats should be sampled in proportion to the availability of each habitat type in the site, while ensuring that all potentially productive habitats are represented in the sample. Surveying the site before sampling will allow the sampler to develop a plan that meets the objective of appropriately representing habitat types in the sample. Sampling procedures that should be used in each habitat type are described below.

In a riffle, start at the downstream edge and place the net firmly in the substrate. Rub by hand any large stones and sticks from within the one foot square area to dislodge any organisms that may be clinging to these substrates. Place these rubbed substrates off to the side. Aggressively disturb the substrate with hand and/or foot. Sampling typically disturbs riffle habitat about 5 to 8 cm below the substrate surface. Rub by hand any large sticks and/or stones from within the disturbed area to dislodge any organisms that may be clinging to these substrates. Repeat this process near the upstream edge of the riffle. Repeat as necessary until the desired number of square feet has been sampled. Samples should be taken from the range of substrate types and velocities found within the riffle to best represent the community of benthic macroinvertebrates living within the riffle.

Log and snag substrates should be rubbed by hand or with a small brush. The D-net should be positioned with the stream current flowing into the net as the logs or snag substrates are rubbed.

The D-net should be used in a jabbing or sweeping motion to dislodge organisms from root mats, submerged macrophytes, or other habitats. Kicking the habitat prior to jabbing may also be done as needed to dislodge organisms. In soft substrates the net motion should be gentler to minimize the collection of detritus. In all cases the D-net should be placed downstream of the sampled substrate following jabbing and sweeping to make sure that dislodged organisms are carried into the net.

In some rare cases (e.g., some large 3rd and 4th order streams), a sufficient amount of potentially productive habitat may not be present within the 75 m site to collect a 20 ft2 sample. If this is the case, moving out of the sample site in an upstream direction to find habitat that can be sampled using a D-net is permissible. This should only be done if it is not possible to collect a sufficient sample within the 75 m site. If sampling is conducted upstream of the 75 m site, a description of the habitats sampled and distance from the upstream end of the 75 m must be recorded in the comments section of the Spring Index Period Data Sheet.

When a complete 20 ft2 sample has been obtained, or when the D-net becomes filled to the point that water does not pass easily through it, the net should be washed into a sieve bucket that is partially submerged and in a shallow portion of a run or pool. While the sample is in the sieve bucket, all large stones (i.e., those greater than 3 cm in diameter), debris, leaves, etc., should be carefully washed, inspected for organisms, and discarded. If necessary, use forceps to remove any animals remaining on the net. All vertebrates (e.g., herpetofauna and fish) should be removed from the sieve bucket at this time. All crayfish should remain in the sample regardless of size. To remove fine sediments from the sample, the sieve bucket may be

gently "slapped" against the stream water surface and very slowly rotated while the bottom of the bucket is submerged. Do not rotate the sieve bucket quickly during this process, as this action may damage many soft-bodied macroinvertebrates potentially rendering them unidentifiable. After processing the sample in the sieve bucket, the benthic net should be rinsed carefully in stream water to make sure that no benthic macroinvertebrates remain that may be transported to the next sample site.

Any unionid mussel or Corbicula incidentally encountered during the Spring Index Period should be recorded on the Spring Faunal Data Sheet. If live mussels are collected in the D-net during benthic macroinvertebrate sampling, they should be placed as closely as possible to where they were collected, or into the appropriate habitat if unsure where the specimen was collected. The mussel should be gently placed partway into the substrate with the anterior end pointing down. Corbicula should remain in the benthic sample.

4. **Preservation.** The processed composite sample should be transferred from the sieve bucket to an externally labeled sample bucket and preserved in 95% ethanol. Place the internal label atop the sample material and ensure that the lid to the sample bucket is tight. Gently mix the sample material and preservative and ready the sample for transport.

5. **Delivery to Laboratory.** A Benthic Macroinvertebrate Chain-of-Custody Sheet (Appendix G) must accompany all samples taken to the benthic macroinvertebrate identification laboratory, which includes the sample identification codes for all samples being delivered, sampler name, date, and a signature from a laboratory representative upon transfer of samples to the laboratory.

   During the spring visit, record in the comments section of the spring data sheet any herpetofauna (positive identifications only) observed or heard at the site, including those released from the sieve bucket during benthic macroinvertebrate processing. Maintain as vouchers any species not previously collected from the basin being sampled.

6. **Archiving.** MBSS benthic macroinvertebrate sample sortates are kept for five years. After this time, the sample material is discarded. Benthic macroinvertebrate subsamples are kept as archives in perpetuity.

5.13.2 Fish Sampling

The objectives of fish sampling for the MBSS are to assess the ecological integrity, fishability, and biodiversity in the non-tidal, flowing waters of Maryland. Double-pass electrofishing of 75 m stream sites is used to collect the information needed to meet these objectives. MBSS electrofishing occurs only during the Summer Index Period (June-September). This time period was chosen to characterize fish communities during the low flow period. Sampling during this period is also advantageous because spawning effects are minimized, temperatures are conducive to wading and water contact, and capture efficiency using electrofishing is typically best when streams are relatively low and warm.

During Round One, each individual fish (up to 100 individuals of each species per site) was inspected for external anomalies. Sites that were sampled during Round One and are being repeat sampled during Round Four will have up to 100 individuals of each species per site inspected individually for external anomalies. Please see number 18 below.

1. **Electrofishing Safety.** All persons conducting electrofishing should be familiar with chapter 3.2.6, in which hazards and procedures for minimizing risk for electric shock, prior to conducting electrofishing are described.

2. **Sampleability.** Prior to conducting electrofishing, the crew leader (with input from other crew members) must determine if the site can be sampled safely and effectively. Electrofishing can only be conducted

safely if the site being sampled can be waded. If the depth or current velocity precludes safe wading, then the site should be considered unsafe for electrofishing. However, where the margins of deep areas can be safely waded and fish can be effectively captured (e.g., using long handled dip nets and anodes), as long as all other sampleability considerations are met, sampling should occur. The most predominant effective sampleability consideration is water visibility. Effective MBSS electrofishing cannot occur in water that is turbid. All areas of the stream bottom must be visible. The only exception to the visibility consideration is a stream that is stained dark from natural organic sources (tannins leached from leaves; blackwater streams). Although sampling can occur in blackwater streams when visibility is relatively limited due to a natural cause, sampling should not occur in a blackwater stream that is also turbid. Whether or not the entire stream bottom is clearly visible in all portions of the site is recorded on the MBSS Fish Data Sheet.

In addition to turbidity and tannic water, overhanging vegetation (especially multiflora rose) may prohibit clear visibility of (and often access to) the stream and habitats that are to be sampled. Provided proper authorization from the landowner has been acquired, vegetation that substantially limits electrofishing should be cleared prior to electrofishing. Block nets should be put in place prior to commencing clearing (or as early as possible during the clearing process) so that fishes do not escape from the site during clearing. Note that when rating shading on the MBSS Summer Habitat Assessment Data Sheet shading that was present before clearing should be recorded.

Other situations that may preclude sampling include dry streams, marshes with no defined channel, impoundments or beaver dams that are too deep to sample, tidally influenced streams, and areas where landowner permission is denied. If a stream is unsampleable (typically due to depth, velocity, or turbidity) during the early part of the Summer Index Period or following a rain event, the stream should be visited later in the Index Period or during a drier period to re-assess sampleability. If (upon return visits) the stream is found to be continuously too deep, fast, or turbid to sample, then the appropriate code for unsampleability (found on the MBSS Summer Index Period Data Sheet) should be recorded on the Summer Index Period Data Sheet.

3. **Sampling Considerations.** The width of the stream, number of anodes needed to effectively electrofish, and any other fish sampling considerations should have been recorded on the Spring Index Period Data Sheet during spring sampling. In cases where spring sampling is not being conducted, site reconnaissance is recommended prior to the electrofishing visit to determine the number of anodes and length of block nets needed, as well as any other fish sampling considerations.

4. **Number of Anodes.** The appropriate number of anodes to cover the entire width of the MBSS site must be used. In all cases this number of anodes is at least one for every three meters of stream width. More than this number may be necessary depending on the amount of habitat available within the stream site, deep areas, or other reasons to be determined by the Crew Leader. All anodes used by MBSS sampling crews are outfitted with ¼" mesh netting to facilitate fish capture. The netting on the anodes should not have any holes or tears greater than ¼". As more than one anode can be used for each electrofishing unit, the number of anodes/unit is recorded on the MBSS Fish Data Sheet.

5. **Dip Nets.** At sites narrow enough to be sampled using only one anode, at least one dip net should accompany the anode for the length of the site. At wider sites, a minimum of one dip net for every two anodes should be used. Dip nets used by MBSS sampling crews have ¼" mesh and should not have any holes or tears greater than ¼". Fishes must be transferred from dip nets to buckets, live cars, or other appropriate storage containers immediately upon capture to limit, as much as possible, stress to each individual fish that is captured.

6. **Barge.** In large, deep, streams it may be deemed necessary by the crew leader to use a floating barge shocker to ensure effective capture of fishes.

7. **Block Nets.** MBSS sampling of fishes requires the use of block nets. Block nets for MBSS sampling should have ¼" or smaller mesh, be completely free of holes or tears larger than ¼" and be long enough to block the entire width of the stream perpendicular to the flow and be high enough to reach from the bottom to above the surface of the stream. Block nets should be placed at the 0 m and 75 m ends of the MBSS site, so as to effectively prohibit the escape of fishes from within the site and to prohibit entry of additional fishes from outside the site. Any tributaries or seeps entering the site that will not be sampled must also be blocked with block nets to prohibit the movement of fish in or out of the site. In braided streams, all braids should be blocked at the 0 m and 75 m locations and all braids should be sampled.

   If the MBSS site includes a culvert that is too small to sample through, block nets should be used to isolate the culvert from the site. The length of the culvert (not the width of the pipe) should then be added to the upstream or downstream end of the site so that the sampled section of stream is a total of 75 m long.

   Although block nets are typically outfitted with small lead weights on the bottom end, these weights are typically insufficient to keep fishes from swimming under the net (especially eels and small benthic species). Therefore, it is necessary to use rocks, stakes, or other objects to anchor the bottom of the net to the stream bottom. Like the lead weights on the bottom of the block net, the top of the block net is also typically outfitted with floats. These floats, however, are typically not sufficient to keep the entire top of the block net above the water's surface, which may permit the escape of small fishes or fishes that can readily jump out of the water. To prohibit the escape of these fishes, it is necessary to lift the top of the block net out of the water and prop it with sticks, rods, or other devices. In most streams, ropes will be needed to anchor the sides of the block nets so that they are not dislodged by the stream current or by floating debris during electrofishing.

8. **Fish Movement.** Prior to and during the installation of block nets care must be taken to ensure that fishes are not chased out of or into the MBSS site. Any observed movement of fishes in or out of the site should be noted on the MBSS Fish Data Sheet. Disturbing the area within and upstream of the site should also be avoided, to the extent possible, prior to electrofishing so that visibility is not affected by resulting turbidity.

9. **Appropriate Voltage.** The output voltage of the electrofishing unit should be adjusted to ensure that fishes are being captured effectively. Proper adjustments of electricity output will vary according to the varying conductivity of the water in different streams. The conductivity should be used as a guide to determine the approximate voltage and frequency to be used. In addition, most electrofishers are equipped with a signal that can be used to guide the adjustment of these settings. Regardless of the conductivity and any signals that the electrofisher provides, testing of the electrofisher's effectiveness downstream of the MBSS site, prior to use in the site, should be conducted, as this is the best way to definitively be sure that the electrofisher is being effective. Effective electrofishing stuns small and large fishes without causing mortality.

10. **Crew Requirements.** All persons participating in electrofishing must wear watertight chest waders. In rocky bottom streams, sticky rubber soles, boot chains, or other appropriate devices must be used to limit slipping on potentially slick substrates. Polarized sunglasses should also be worn to reduce glare and thereby improve capture efficiency. Under cloudy or lower light conditions, amber-lensed glasses should be worn, while green-lensed or brown-lensed glasses are appropriate under sunny conditions. The use of rubber gloves is highly recommended due to the danger of electric shock that could occur from contact with water being sampled.

11. **Time.** The seconds of electrofishing for each unit being used for sampling should be monitored and recorded for each of the two electrofishing passes. On the MBSS Fish Data Sheet, the time in seconds is recorded as the time each unit reads at the beginning of the first electrofishing pass, at the beginning of the second electrofishing pass, and at the end of the second pass.

12. **Fish Sampling.** MBSS electrofishing begins at the downstream block net. The entire site is thoroughly electrofished, bank to bank, including backwater areas, sloughs, and shallows, making an equal attempt to capture every fish observed. An exception is that fish too small to be retained by dip nets (body length less than 30 mm) need not be collected. When necessary to ensure capture of fish, the operator of the electrofishing unit should use the net on the anode ring. For the MBSS, continuous rather than intermittent electrofishing is used to avoid bias introduced by selective placement of the electrode and reduce sampling mortality.

    All captured fish are placed into buckets, live cars, or other appropriate storage containers immediately upon capture to limit, as much as possible, stress to each individual fish that is captured. Providing water flow through live cars and bubblers will substantially increase survival of collected fishes compared to using closed systems without bubblers. Care should be taken to avoid electrofishing near any flow-through containers as the fishes in these containers will be affected by the electricity.

    In fast water or where visibility is reduced, dip netters should place nets on the stream bottom to increase the probability of capturing bottom dwelling individuals that may be difficult to see. Particular attention should be given to capturing small benthic fishes (darters, sculpins, and madtoms).

13. **Block Net Check.** Upon completion of each electrofishing pass, the entire downstream block net must be examined for fishes. It is important to make sure that the downstream block net still effectively blocks the movement of fishes after checking it following first pass electrofishing. This may require the removal of debris that has accumulated during the first pass.

14. **Delaying Second Pass.** If water clarity in the site is reduced because of substrate disturbance during the first pass, second pass electrofishing must be delayed up to one hour until visibility improves to the point that visibility is similar to what it was during the first pass. If 2nd pass visibility is poorer than 1st pass visibility, it should be noted on the Fish Data Sheet.

15. **Equal Effort.** To ensure consistency among MBSS sampling crews, it is important to use the same sampling effort on the second pass as was expended for the first pass. This requires that all of the same habitat that was sampled during the first pass be sampled on the second pass. Therefore, the entire site should be electrofished on the second pass. The number of units, netters, and anodes should also be the same during both electrofishing passes.

16. **Biomass.** Fishes are weighed in aggregate to the nearest 10 grams separately for the first and second electrofishing pass. Only fishes should be weighed. Other organisms, rocks, sticks, leaves and other debris must be removed prior to weighing. Aggregate fish biomass is recorded at the bottom of the MBSS fish Data Sheet.

17. **Counting Fishes.** All fishes captured are identified to species and enumerated. The numbers of fish by species are recorded separately for the first and second pass. A "Fish Crib Sheet" is provided on Appendix H to aid in counting by species.

    All individuals not clearly identifiable to species should be retained for later inspection in the laboratory. The number of individuals retained should be indicated on the MBSS Fish Data Sheet. Retention of all specimens which cannot be positively identified is mandatory. Specimens for preservation should be promptly placed into plastic jars filled with a 10% buffered formalin solution. After a minimum of five days, but no more than a month in formalin, the specimens should be soaked for 24-48 hours in water, after which they can be transferred to 70% ethanol solution. Individuals >160 mm should be slit on the lower abdomen of the RIGHT side prior to preservation in formalin. All specimen jars should be labeled with inside labels specifying the date, site number, and name of collector. An example of the MBSS Voucher

Specimen Label is included on Appendix I. The MBSS fish key should be used as needed for positive verification during field identifications. Only persons who have passed the MBSS fish taxonomy test should identify fishes to species in the field.

18. **Unusual Anomalies.** For each species, if any unusual occurrences of anomalies are observed it should be recorded with a Y on the "Unusual Anomalies" section of the MBSS Fish Data Sheet. If unusual anomalies are not observed, an N should be entered in this section of the data sheet. Unusual anomalies can include, but are not limited to, excessive black spot or black spot on an atypical species, multiple skeletal deformities, fin erosion, lesions, tumors, fungus, discoloration, excessive external parasites, or other unusual appearance. Any other comments, by species, that may be important in fish data analysis or interpretation can also be entered on the MBSS Fish Data Sheet.

    For sites previously sampled during Round One, up to 100 individual fishes of each species should be inspected for any external anomalies. The number of individuals, by species, with any external anomaly, by anomaly type, should be recorded on the MBSS Fish Data Sheet. The following codes should be used on the data sheet for anomalies:

    DI = Discoloration
    CL=Fin Cloudiness
    BL=Black Spot
    UL=Ulcertations/Lesions
    FI= Fin Erosion
    DV=Deformities of the Vertebral Column
    AN=Swelling of the Anus
    RE=Red Spot
    OT=Other (define in comments on the data sheet)
    FD=Fin Deformed or Missing
    IK=Ich
    LE=Leeches
    EC=Eye Cloudiness
    PO=Exopthalmia (pop eye)
    NO= Eye Missing

    HM= Hemorrhaging
    CL= Raised Scales
    GR=Growths/Cysts
    EP= Visible External Parasites
    FU= Fungus
    DM=Deformities of the Mandible
    SC= Scale Deformities
    HK=Hooking Injury
    BS=Body Shape (NOT BLACK SPOT)
    CT=Cut
    AW=Anchor Worm
    FU=Fungus
    HE=Eye Hemorrhage
    OR=Depression Into the Orbits
    CA= Cataract

19. **Voucher Specimens.** For the Round Four MBSS, DNR field crews will be required to maintain voucher collections. However, photographic vouchers will be acceptable in lieu of preserved specimens, provided the features that need to be seen to correctly identify the specimen are clearly visible in the photograph. Photographs of at least five specimens of each fish, reptile, amphibian, and crayfish species encountered during Round Four (as long as five were collected) should be photographed. In addition, any rare, threatened, or endangered species encountered should be photographed, as long as the photograph can be taken without causing any harm to the specimen. The Maryland Department of Natural Resources, Monitoring and Non-Tidal Assessment Division will keep a voucher library of all photographs taken during MBSS sampling. With the exception of rare, threatened, or endangered species, specimens for which photographs cannot be used to verify identifications should be preserved for verification. Photographs will be reviewed by an expert in taxonomy for each taxonomic group and results will be kept on record. Each species photograph should include a label with the date and site identification.

20. **Taxonomic experts** (or a designee assigned by the taxonomic expert) will also audit field identification of organisms. Field audits will be conducted by taxonomic experts (or designee) at a minimum of one site per crew.

    Release individuals not retained as voucher specimens or for laboratory examination. Extreme care should be taken while holding fish prior to release to reduce stress from handling and crowding. Plenty of oxygenated water should also be supplied by holding fishes in stream flow through live cars and/or using battery-operated bubblers.

21. **Gamefish.** During counting and identification of the fish sample for each pass, each gamefish species collected should be measured to the nearest mm (total length) and recorded on the Gamefish Length Data Sheet. Gamefish species for the MBSS include all bass (*Micropterus spp.*), striped bass (*Morone saxatilis*), trout (*Salmonidae*), walleye (*Sander vitreum*), and pikes and pickerels (*Esocidae*) with the exception of redfin pickerel (*Esox americana*). If visual observations suggest that some individuals may be stocked fish (based on fin wear, fin size, etc.), indicate so in the comments section for that species on the Fish Data Sheet.

5.13.3 Reptile and Amphibian Sampling

Reptile and amphibian sampling has been an integral part of the Maryland Biological Stream Survey since 1994. Reptiles and amphibians, particularly stream salamanders, have been shown to be excellent indicators of MBSS site conditions. During Round Two, only incidental observations of herpetofauna were recorded for MBSS sites, with no focused search conducted. Round Three emphasized searching for stream salamanders and spending a minimum of fifteen minutes searching available habitat for herpetofauna. In Round Four, stream salamander sampling will follow a protocol shown to adequately support population estimates and use of the stream salamander Index of Biotic Integrity (SS-IBI). Incidental observations will also continue to be recorded. The life-history stage of reptiles and amphibians should also be recorded (egg, larva, or adult).

1. **Incidental Collection.** During Round Four, the full common name of any reptile or amphibian species that is encountered while sampling or accessing MBSS sites, during either spring or summer, should be recorded on the MBSS Spring Fauna Data Sheet or Summer Fauna Data Sheet, respectively.

2. **Stream Salamanders.** Stream salamanders include the following species in the family Plethodontidae; the northern red salamander, eastern mud salamander, northern spring salamander, northern two-lined salamander, long-tailed salamander, northern dusky salamander, Appalachian mountain dusky salamander, and seal salamander. Stream salamander sampling consists of electrofishing in aquatic habitat and bank searches in terrestrial habitat. All stream salamanders found incidentally, during the bank search, or while electrofishing should be counted by species and life history stage (e.g., larva or adult).

    Electrofishing protocols are described in the fish sampling portion of this manual. Salamanders should be collected coincident with fish collection during electrofishing. Bank searches focused on finding stream salamanders will be conducted during the Summer Index Period visit, along both banks of the 75-m site. Although the bank search will be conducted entirely outside the wetted portion of the stream, the bank search should focus on cover objects (including cobbles, small boulders, logs, or other objects) where sufficient moisture is present to support stream salamanders. In streams where seeps or small tributaries enter the stream, these habitats should also be searched to a distance 10 m from the main channel. Cover objects in places that are completely dry have a lower likelihood of supporting salamanders and do not need to be searched. During the search, all suitable cover adjacent to the site should be carefully flipped over and then returned as closely as possible to the original position. In some Maryland streams, such suitable cover can extend several meters from the edge of the stream before the habitat is completely dry and not suitable.

Recognizing that a small proportion of MBSS sites will have high abundances of salamanders, salamander transect searches will be limited to 60 minutes. If all stream salamander habitat is searched in less than 60 minutes, then searching will stop. It is important that both stream banks are searched at a consistent, steady pace. In those unusual cases where all stream salamander habitats (i.e., the cover objects described above) have not been sampled in 60 minutes, the crew should estimate the remaining amount of habitat (including seeps) as a proportion of the entire site. For example, the crew might estimate that of the 100% of stream bank with cover objects, only 80% was searched in 60 minutes and 20% remains after sampling stopped. The percentage of habitat remaining (unsearched) should be recorded on the data sheet. Research has shown that 97% of all stream salamander species are sampled in the first 60 minutes, so the percentage of unsampled, remaining habitat will only be used to improve estimates of salamander abundance.

3. **Sampleability.** Bank searches can be conducted in some streams even when electrofishing cannot (e.g., dry stream channel). However, bank searches may be precluded by certain stream conditions. As with other aspects of MBSS sampling, the sampleability codes provided on Appendix D should be used to indicate sampleability for stream salamanders.

4. **Photographic Vouchers.** Photographs should be taken of any rare, threatened, or endangered species (provided the photograph can be taken without harming the specimen). In addition, voucher photographs of at least five individuals (provided at least 5 individuals are encountered) of each species encountered should be taken to verify proper identification in the field. Photographs should show the anatomical features that are necessary for proper taxonomic identification and files for digital photographs should include the MBSS site identification. The best photographs have the site identification in the photograph, with the specimen. The Department of Natural Resources, Monitoring and Non-Tidal Assessment Division will keep a photographic voucher collection for reptiles and amphibians. It is not necessary to preserve any reptile or amphibian specimens during the Round Four MBSS.

5. **Taxonomic Identification.** Only those members of the field sampling crew who have passed the reptile and amphibian taxonomy test should perform reptile and amphibian identification for the crew. The Key to the Reptiles and Amphibians of Maryland can be consulted to help with identifications.

6. **Frog Calls.** A frog call test will be administered during the Spring Index Period training. Field crew members who pass the frog call test can identify frogs as present in the vicinity of MBSS sites based on hearing the frog call. Frogs that are heard calling can be recorded on the Spring Index Period Data Sheet during spring sampling and on the Summer Index Period Data Sheet during summer sampling.

7. **Handling Care and precautions.** Live specimens that will be released should be handled as little as possible, while still ensuring sufficient observation to obtain accurate taxonomic identification. Animals should be released as closely as possible to where they were captured. If an animal was found under a cover object, the object should be returned to its original position and the animal should be placed next to the object and allowed to return to underneath the object on its own.

5.13.4 Crayfish Sampling

1. **Sampleability.** If the site can be safely accessed and with landowner permission, sampling for crayfishes during the summer index period should be conducted. This includes sampling in dry streams. During the spring and summer index period incidental findings should also be recorded.

2. **Stream Crayfishes.** An attempt should be made to capture all crayfishes encountered during each electrofishing pass. Most stream-dwelling crayfishes are primarily nocturnal and reside in shallow burrows under stream substrate (e.g. cobbles, boulders, woody debris) during the day. Effort should be made during each electrofishing pass to overturn or disturb these habitats to optimize the number crayfishes captured. All captured crayfishes are placed into buckets, live cars, or other appropriate storage containers

immediately upon capture to limit, as much as possible, stress to each individual crayfish that is captured. Upon the completion of each pass, the downstream blocknet is checked for crayfishes. Identify and enumerate all adult (>15 mm carapace length) crayfishes caught during the first and second electrofishing pass. The full scientific (Latin) name of each species and the number collected during each pass are recorded in the crayfish section of the Summer Index Period Data Sheet.

3. **Burrows.** The presence of crayfish burrows along stream banks or within the floodplain adjacent to the MBSS site is recorded on the datasheet. The abundance of burrows is recorded as (P): Present, (A): Absent, or (E): Extensive. If time allows, an attempt should be made to excavate crayfish burrows to identify the burrowing species.

4. **Taxonomic Identification.** Only those members of the field sampling crew who have passed the crayfish taxonomy test should perform crayfish identification for the crew. The Key to the Crayfishes of Maryland can be consulted to help with identifications.

5. **Crayfish Vouchers.** All specimens that cannot be positively identified in the field should be retained for further inspection by a regional crayfish expert. Field identification of *Procambarus acutus* and *P. zonangulus* is difficult and unreliable. All specimens of these two species should be recorded on the datasheet as *Procambarus acutus*/*zonangulus* and vouchered to allow for species-level identification in the laboratory. Retention of all specimens which cannot be positively identified is mandatory. For Round Four MBSS, one to five individuals of each species collected by each MBSS field crew should be retained so that taxonomic identifications can be verified. The preservation of Form I males is preferred. However, females and small males should be vouchered in the absence of Form I males. Specimens for preservation should be promptly placed into plastic jars filled with 70% ethanol solution. Label all specimen jars with an inside label specifying the date, site name, and name of collector. Release all crayfishes not retained for vouchers.

5.13.5 Mussel Sampling

Any freshwater mussel (Family Unionidae), Dreissenidae, or Asiatic clams (Corbicula) that are observed while sampling MBSS sites should be identified to species with their scientific names recorded. During the Summer Index Period, suitable bivalve habitats within the sampling segment should be searched, with part of this effort focused on searching the stream bank for shells and animal middens. This can be conducted in conjunction with the herpetofauna search, but should consist of at least 15 minutes of effort. Live specimens that are encountered should be identified in the field, and then immediately returned as closely as possible to where they were collected. The mussel should be gently placed partway into the substrate with the anterior end pointing down. The species encountered and whether they were live (L) or dead (D) should be noted on the Summer Fauna Data Sheet in the appropriate area. A check box is provided to record the apparent absence of mussels from the site. If no Corbicula are encountered, it should be noted on the data sheet as none "N" in the section of the data sheet designated for recording information about Corbicula.

Any unionid mussel or Corbicula incidentally encountered during the Spring Index Period should be recorded on the Spring Faunal Data Sheet. If live mussels are collected in the D-net during benthic macroinvertebrate sampling, they should be placed as closely as possible to where they were collected, or into the appropriate habitat if unsure where the specimen was collected. The mussel should be gently placed partway into the substrate with the anterior end pointing down.

No live freshwater mussels should be vouchered. Digital pictures should be taken of live specimens, for which the identification is uncertain provided that the photographs clearly show characters necessary to confirm the identification. At a minimum, photodocumentation will clearly show a lateral and a dorsal aspect of each specimen. Additional characters that may prove beneficial to identification include umbo/beak sculpture and posterior slope. Placing the specimen against a light-colored background for the picture may help produce a

clear photograph. Pictures should be forwarded to a mussel expert for confirmation. Valves from as many dead specimens as practical for which the identification is uncertain should be retained. Valves collected from a single site can be placed in one zip-lock bag with a voucher label containing site name, date, and collector. Voucher shells should be cleaned of all debris with a soft brush (e.g. toothbrush) in water before sending to a taxonomic expert for verification.

While it is rare to find evidence of mussels in streams that are dry when visited during the Summer Index Period, they have the ability to withstand short periods of drought. Therefore, mussel sampling can be conducted in streams with standing pools or streams that have become dry.

5.13.6 Invasive Plants

The full common name of invasive plants observed at each MBSS site is recorded during the Summer Index Period. The common names of any invasive plant species observed within view of the MBSS site should be recorded. However, the riparian area within five meters of the stream on each bank should be thoroughly searched. The abundance of each invasive plant found is recorded as present (P) or Extensive (E). Only those members of the field sampling crew who have passed the exotic plant taxonomy test should perform invasive plant identification for the crew.

Appendices

Appendix A: MBSS Round Four Design Summary

Appendix B: Fish Fixation and Preservation Procedures

Appendix C: Equipment Lists

Appendix D: Codes and Abbreviations

Appendix E: MBSS Stream Habitat Assessment Guidance Sheet

Appendix F: Species Names

Appendix G: Chain of Custody Forms

Appendix H: MBSS Voucher Label

Appendix I: MBSS Summer Data Sheets

Appendix J: MBSS Spring Data Sheets

Appendix K: MBSS Geomorphology Data Sheets

Appendix A: MBSS Round Four Design Summary

1. *Introduction*

The Maryland Department of Natural Resources (DNR) will begin Round Four (R4) of the Maryland Biological Stream Survey (MBSS) in the spring of 2014. The MBSS is a stratified-random, probabilistic survey that provides essential information on the ecological condition of Maryland streams for the State's natural resource decision makers. The primary goal for R4 of the MBSS is to document changes in stream conditions over time. R4 will also provide status information at the large watershed (basin) and statewide scales.

2. *Survey Design*

R4 will include sampling over the 5-year period of 2014-2018. The R4 sites will be sites previously sampled during Rounds One and Two of the MBSS. These "repeat" R4 sites will be randomly selected from the originally randomly selected sites in Round One (R1) during 1995-1997 and Round Two (R2) during 2000-2004. The R1 repeat sites will be sampled in 2015-2017, following the same annual allocation of sites so that the period between sampling will be 20 years for each site. The R2 repeat sites will be sampled 2014-2018, following the same annual allocation of sites so that the period between sampling will be 14 years for each site.

The following number of repeat sites will be sampled each year using the design of the original round:

Year	R1 Sites	R2 Sites	R4 Core Sites	Targeted R4 Sites*
2014	--	48 sites in 16 PSUs	48	7
2015	54 sites in 6 basins	51 sites in 17 PSUs	105	7
2016	54 sites in 6 basins	51 sites in 17 PSUs	105	TBD
2017	45 sites in 5 basins	51 sites in 17 PSUs	96	TBD
2018	--	51 sites in 17 PSUs	51	TBD
Total			405	TBD

*This final number of sites has not been determined for certain. The final number will most likely consist of repeat sampling all MBSS sites that were sampled during R1 and R2 in certain "special interest" watersheds. An example of this targeted sampling is Mattawoman Creek – described below. Other targeted sampling sites and watersheds will be added to each year's effort after determination of the priority "special interest" watersheds.

This totals 153 sites from R1 and 252 sites from R2 for 405 core repeat sites in R4.

The sample design for the repeat sites from R1 will follow the R1 design of random selection of stream reaches from strata defined by basin and stream order, specifically equal probability of selection in 1st, 2nd, and 3rd order streams within each of 17 basins statewide. 3 repeat sites will be randomly selected from the R1 sites in each stratum, where possible (some strata have low numbers of R1 sites to select from).

The sample design for the repeat sites from R2 will follow the R2 design of random selection of stream reaches from Primary Sampling Units (PSUs) that generally equate to Maryland 8-digit watersheds (which are combined when they are small to make 84 PSUs statewide). three repeat sites will be randomly selected from the R2 sites in each PSU.

Given that R1 and R2 used different sample designs and stream network map scales (R1 sites were selected from the 1:250,000-scale map and R2 sites were selected from the 1:100,000-scale map), the inter-round comparison of repeated samples will be conducted separately for each round. The R1 comparison will be a 20-year comparison; the R2 comparison will be a 14-year comparison. Each of these inter-round comparisons will have the ability to detect a change of 0.20-0.25 BIBI units with 80% probability.

3. *Watershed Intensification*

To increase the density of samples in certain watershed of special interest, all sites sampled in these watersheds in R1 and R2 will be repeat sampled in R4. For example, the 7 sites sampled for R1 in the Mattawoman Creek watershed during 1995 (some of these may be included in the 3 repeat sites for the R4 Lower Potomac sampling) will be repeat sampled in 2015, while all 10 sites sampled for R2 in 2000 will be repeat sampled in 2014 (these 10 sites include the 3 core R4 samples and 7 samples for additional density). The complete list of special interest watersheds will be reviewed each year during Round Four.

4. *Bookkeeping and Logistics*

It should be noted that some of the repeat sites selected randomly from R1 or R2 may have been designated as MBSS sentinel sites and sampled annually since they were first designated. If one or more of these sites are selected randomly for R4, their annual sentinel sampling results in the appropriate year will be included in R4 (a separate record denoting membership in R4 should be created in the database).

To obtain site access permissions, landowners for the first 10 sites per strata randomly selected for R4 from R1 and R2 will be identified and letters requesting permission will be mailed. The goal will be to obtain permissions for twice the number of sites to be sampled per stratum (i.e., 2x3=6) and to provide them to DNR.

5. *Additions and Modification to Sampling and Assessment Protocols*

The R4 MBSS will sample the same parameters sampled during R3, with one addition and one modification. Geomorphological assessments will be added. This will consist of a rapid assessment of geomorphology at randomly-selected R4 sampling sites (exact protocols to be determined) and detailed geomorphological assessments (consisting of cross-sections, longitudinal profiles, and pebble counts) at all MBSS Sentinel Sites. By increasing the time and area searched, the stream salamander sampling protocol has been modified from R3 to provide a greater likelihood of collecting stream salamanders. The goal is to support the development of a salamander index of biotic integrity, a biological indication that will be especially useful in small headwater streams with naturally depauperate fish assemblages. More detailed descriptions of these R4 sampling protocols will be included in an updated version of the MBSS sampling manual, scheduled for completion by February 2014.

Appendix B: Fish Fixation and Preservation Procedures

Fixation disables proteins such as enzymes that cause cell lysis and thus stops the cellular degradation. However, fixation also breaks up DNA so is not good for genetic samples.

* fixation - use 10% formalin for juveniles and adults; 5% for larvae. Formaldehyde gas in 40% aqueous solution can be diluted 9:1 with water to make 10% formalin. The volume of fishes in the sample themselves must be taken into account upon fixation, so the initial solution should be stronger than 10% (e.g., 20%). Make a one-inch cut in larger specimens (>150 mm) on the right hand side of the fish above and in front of anus. **Put fish in a large screw-top container with a fluid to fish ratio of at least 2:1 and ideally 5:1.** Try to get fish to lie straight - laying the bottle on its side for a while helps. You can begin transfer of fish to a storage fluid after at least 48 hours in formalin.

Storage fluids are used to maintain fixed or preserved fish for long periods. For fixed fish, formalin is rinsed off by soaking specimens in water for at least two days and up to one week. During this period, **water should be changed at least four times. Note that formalin and formalin-fixed specimens are considered toxic wastes and must be disposed of properly.** Decanted formalin can be saved for reuse in proper concentration. Rinsed fish are then transferred into 70% ethanol or 45-50% isopropanol.

Preservation tries to stop tissue degradation by removing liquid water. This can happen with freezing, using salts, or alcohol. The typical field approach is to use 95% ethanol, which allows recovery of DNA, but the fish's morphology is altered making them difficult to work with for morphology.

* preservation - use 95% ethanol. Make a one-inch cut in larger specimens (>150 mm) on the right hand side of the fish above and in front of anus. **Put fish in a large container with a fluid to fish ratio of about 5:1.** For best results, decant after 24 hrs and replace with fresh ethanol. Preserved fish are usually stored in 70-90% ethanol.

Detailed instructions for fixation and preservation can be found in Kelsh and Shields (1996).

References

Maryland Natural Heritage Program. 2010. Rare, threatened, and endangered animals of Maryland. Maryland Department of Natural Resources, Wildlife and Heritage Service, Annapolis, MD.

Kelsh, S.W. and B. Shields. 1996. Care and handling of sampled organisms. Pages 121-144 in B.R. Murphy and D.W. Willis, editors. Fisheries techniques, 2nd edition. American Fisheries Society, Bethesda, MD.

Stauffer, J.R., J. Karish, and T.D. Stecko. 2001. Guidelines for using digital photos as fish vouchers for Pennsylvania Fishes. The Pennsylvania State University and Natural Park Service

Appendix C: Equipment Lists

Spring Index Period Equipment List This table lists the equipment needed to complete sampling for all variables for which MBSS field sampling is conducted during the Spring Index Period.	
MBSS Sampling Manual	Chest waders
Road maps and itinerary	Wader repair kit
Site list/maps	G.P.S. unit
Spring Habitat Data Sheets	Compass
Spring Index Period Data Sheets	Ice
Vernal Pool Data Sheets	Bubble wrap, packing material, packing tape
Permanent markers	Clear label tape
Pencils	Deionized water for blanks
Taxonomic Keys (reptiles and amphibians, crayfish, freshwater mussels)	Water quality sample bottles- 1 liter
Machete or other clearing tools	Water quality sample bottles- 500 mL
Digital camera	Syringes and valves
First aid kit	Tripod, level, and stadia
Spray paint	Pre-printed adhesive outside labels and inside labels
Flagging	Spare batteries
100 m measuring tape	Chain-of-custody forms
540 micron mesh D-net (frame dimensions 12” W x 10” H)	Ziplock bags
Spare net bag for D-net	Pump sprayer or decontamination solution container
540 micron sieve bucket	Rinse water
Benthic sample containers (86 oz or larger)	Decontamination solution – 10% bleach or Virkon
EtOH (2 liters per site)	Extra drinking water
Foul weather gear	Backpack
Small cooler for transporting water samples from site to vehicle	Large cooler for keeping samples cool after collection and for shipping to laboratory
Temperature loggers	

Summer Index Period Equipment List

This table lists the equipment needed to complete sampling for all variables for which MBSS field sampling is conducted during the Summer Index Period.

MBSS sampling manual	Flowmeter and staff gauge
Road maps	Spring or electronic scale
Site list and site maps	Calibration weights
Summer Index Period Data Sheets	Pruning tool
Fish Data Sheets	Backpack electrofishing Unit(s)
Summer Habitat Data Sheets	Anode ring probe(s) (fitted with 3/16" mesh netting);
Gamefish Length Data Sheets	Electrofishing batteries
Summer Photo Data Sheets	Spare netting/cable ties for anode ring nets
Habitat Guidance Sheet	25 liter buckets
Clipboards	Dip nets
Pencils	Block nets
Sample jars	Live cars
Pre-printed voucher labels	Machete
Taxonomic keys (reptiles and amphibians; fish, freshwater mussels, crayfishes)	Calibration log
Voucher lists	Waders and wading boots
Preservatives (alcohol and formalin)	Cellular phone
100 m measuring tape	Backpacks
Flagging	Measuring board
Digital camera	Meter sticks
G.P.S. unit	Tool box
Compass	Wader repair kit
Disinfectant lotion	Polarized Glasses
Drinking water	Pump sprayer or decontamination solution container
First aid kit	Rinse water
Foul weather gear	Decontamination solution – 10% bleach or Virkon
Extra drinking water	

Appendix D: Codes and Abbreviations

Watershed Abbreviation

Aberdeen Proving Ground	ABPG
Anacostia River	ANAC
Antietam Creek	ANTI
Assawoman Bay	ASSA
Atkisson Reservoir	ATKI
Atlantic Ocean	ATLA
Back River	BACK
Back Creek	BACR
Baltimore Harbor	BALT
Big Annemessex River	BANN
Big Elk Creek	BELK
Bird River	BIRD
Bodkin Creek	BODK
Bohemia River	BOHE
Breton Bay	BRET
Brighton Dam	BRIG
Broad Creek	BROA
Bush River	BUSH
Bynum Run	BYNU
Cabin John Creek	CABJ
Casselman River	CASS
Catoctin Creek	CATO
Conowingo Dam Susquehanna R	CDAM
Chincoteague Bay	CHIN
Christina River	CHRI
Conewago Creek	COCR
Conococheague	CONO
Corsica River	CORS
Deep Creek Lake	DCRL
Deer Creek	DEER
Dividing Creek	DIVI
Double Pipe Creek	DOUB
Eastern Bay	EAST
Evitts Creek	EVIT
Fifteen Mile Creek	FIMI
Fishing Bay	FISH
Furnace Bay	FURN
Georges Creek	GEOR
Gilbert Swamp	GILB
Gunpowder River	GUNP
Gwynns Falls	GWYN
Honga River	HONG
Isle of Wight Bay	ISLE
Jones Falls	JONE
Kent Island Bay	KEIS
Kent Narrows	KENA
Langford Creek	LANG
Little Conococheague	LCON
Liberty Reservoir	LIBE
Little Choptank	LICK
Little Elk Creek	LIEL
Little Gunpowder Falls	LIGU
Licking Creek	LIKG
Lower Monocacy River	LMON
Loch Raven Reservoir	LOCH
Lower Choptank	LOCK
Lower Chester River	LOCR
Lower Elk River	LOEL
Lower Gunpowder Falls	LOGU
Lower Pocomoke River	LOPC
Lower Wicomico	LOWI
Little Patuxent River	LPAX
Lower Susquehanna	LSUS
Little Tonoloway	LTON
Lower Chesapeake Bay	LWCH
Lower Winters Run	LWINT
Little Youghiogheny	LYOU
Magothy River	MAGO
Manokin River	MANO
Marsh Run	MARS
Marshyhope Creek	MACK
Mattawoman Creek	MATT
Middle Chesapeake Bay	MDCH
Middle Chester River	MICR
Middle River-Browns	MIDD
Miles River	MILE
Monie Bay	MONI
Middle Patuxent Rier	MPAX
Nanjemoy Creek	NANJ
Nanticoke River	NANT
Nassawango Creek	NASS
Northeast River	NEAS
Newport Bay	NEWP
Octoraro Creek	OCTO
Oxon Creek	OXON
Patapsco River Lower North Br	PATL
Patuxent River Lower	PAXL
Patuxent River Middle	PAXM
Patuxent River Upper	PAXU
Pocomoke Sound	PCSO
Piscataway Creek	PISC
Potomac AL Co	PRAL
Prettyboy Reservoir	PRET
Potomac River FR Co	PRFR
Potomac River Lower North Br	PRLN
Potomac Lower Tidal	PRLT
Potomac River MO Co	PRMO
Potomac River Middle Tidal	PRMT
Potomac River Upper North Br	PRUN
Potomac Upper Tidal	PRUT
Potomac WA Co	PRWA
Port Tobacco River	PTOB
Rocky Gorge Dam	RKGR
Rock Creek	ROCK
Sassafras River	SASS
Savage River	SAVA
South Branch Patapsco	SBPA
Southeast Creek	SEAS
Seneca Creek	SENE
Severn River	SEVE
Sideling Hill Creek	SIDE
Sinepuxent Bay	SINE
South River	SOUT
St. Clement Bay	STCL
Stillpond-Fairlee	STILL
St. Mary's River	STMA
Swan Creek	SWAN
Tangier Sound	TANG
Tonoloway	TONO
Town Creek	TOWN
Transquaking River	TRAN
Tuckahoe Creek	TUCK
Upper Elk River	UELK
Upper Monocacy River	UMON
Upper Chesapeake Bay	UPCH
Upper Choptank	UPCK
Upper Chester River	UPCR
Upper Pocomoke River	UPPC
West Chesapeake Bay	WCHE
Western Branch	WEBR
West River	WEST
Wicomico River	WICO
Wicomico Creek	WICR
Wills Creek	WILL
Wicomico River Head	WIRH
Wye River	WYER
Youghiogheny River	YOUG
Zekiah Swamp	ZEKI

Vegetation Types

G = Grasses /Forbes
R = Regen Deciduous /Shrubs (<4"DBH)
Y = Young Deciduous (4-12" DBH)
M = Mature Deciduous (12-24" DBH)
O = Old Deciduous (>24" DBH)
A = Regen Coniferous (<4" DBH)
B = Young Coniferous (4-12" DBH)
C = Mature Coniferous (12-24" DBH)
D = Old Coniferous (>24" DBH)
L = Lawn

Riparian Buffer Zone/ Adjacent Land Cover Types

FR = Forest
OF = Old Field
EM = Emergent Vegetation
LN = Mowed Lawn
TG = Tall Grass
LO = Logged Area
SL = Bare Soil
RR = Railroad
PV = Paved Road
PK = Parking Lot/ Industrial/ Commercial
GR = Gravel Road
DI = Dirt Road
PA = Pasture
OR = Orchard
CP = Cropland
HO = Housing

Instream Blockage Codes

DM = Dam
PC = Pipe Culvert
F = Fishway
GW =Gaging Station Weir
G = Gabion
PX = Pipeline Crossing
AC = Arch Culvert
BC = Box Culvert
TG = Tide Gate
(Note: Height is measured in meters from stream surface to water surface above structure)

Sampleability Codes

S = Sampleable
1 = Dry Stream bed
2 = Too Deep
3 = Marsh, no defined channel
4 = Excessive Vegetation
5 = Impoundment
6 = Tidally Influenced
7 = Permission Denied
8 = Unsafe (Describe in Comments)
9 = Beaver
10 = Other

Appendix E: MBSS Stream Habitat Assessment Guidance Sheet

MBSS Stream Habitat Assessment Guidance Sheet				
Habitat Parameter	**Optimal 16-20**	**Sub-Optimal 11-15**	**Marginal 6-10**	**Poor 0-5**
1. Instream Habitat[1]	Greater than 50% of a variety of cobble, boulder, submerged logs, undercut banks, snags, root wads, aquatic plants, or other stable habitat	30-50% of stable habitat. Adequate habitat	10-30% mix of stable habitat. Habitat availability less than desirable	Less than 10% stable habitat. Lack of habitat is obvious
2. Epifaunal Substrate[2]	Preferred substrate abundant, stable, and at full colonization potential (riffles well developed and dominated by cobble; and/or woody debris prevalent, not new, and not transient)	Abund. of cobble with gravel &/or boulders common; or woody debris, aquatic veg., under-cut banks, or other productive surfaces common but not prevalent /suited for full colonization	Large boulders and/or bedrock prevalent; cobble, woody debris, or other preferred surfaces uncommon	Stable substrate lacking; or particles are over 75% surrounded by fine sediment or flocculent material
3. Velocity/Depth Diversity[3]	Slow (<0.3 m/s), deep (≥0.5 m); slow, shallow (<0.5 m); fast (≥0.3 m/s), deep; fast, shallow habitats all present	Only 3 of the 4 habitat categories present	Only 2 of the 4 habitat categories present	Dominated by 1 velocity/depth category (usually pools)
4. Pool/Glide/Eddy Quality[4]	Complex cover/&/or depth ≥1.5m; both deep (≥ .5 m)/shallows (< .2 m) present	Deep (≥0.5 m) areas present; but only moderate cover	Shallows (<0.2 m) prevalent in pool/glide/eddy habitat; little cover	Max depth <0.2 m in pool/glide/eddy habitat; or absent completely
5. Riffle/Run Quality[5]	Riffle/run depth generally >10 cm, with maximum depth greater than 50 cm (maximum score); substrate stable (e.g. cobble, boulder) & variety of current velocities	Riffle/run depth generally 5-10 cm, variety of current velocities	Riffle/run depth generally 1-5 cm; primarily a single current velocity	Riffle/run depth < 1 cm; or riffle/run substrates concreted
6. Embeddedness[6]	Percentage that gravel, cobble, and boulder particles are surrounded by line sediment or flocculent material.			
7. Shading[7]	Percentage of segment that is shaded (duration is considered in scoring). 0% = fully exposed to sunlight all day in summer; 100% = fully and densely shaded all day in summer			
8. Trash Rating[8]	Little or no human refuse visible from stream channel or riparian zone	Refuse present in minor amounts	Refuse present in moderate amounts	Refuse abundant and unsightly

1. **Instream Habitat.** Rated based on perceived value of habitat to the fish community. Within each category, higher scores should be assigned to sites with a variety of habitat types and particle sizes. In addition, higher scores should be assigned to sites with a high degree of hypsographic complexity (uneven bottom). In streams where ferric hydroxide is present, instream habitat scores are not lowered unless the precipitate has changed the gross physical nature of the substrate. In streams where substrate types are favorable but flows are so low that fish are essentially precluded from using the habitat, low scores are assigned. If none of the habitat within a segment is useable by fish, a score of zero is assigned.

2. **Epifaunal Substrate.** Rated based on the amount and variety of hard, stable substrates usable by benthic

macroinvertebrates. Because they inhibit colonization, floculent materials or fine sediments surrounding otherwise good substrates are assigned low scores. Scores are also reduced when substrates are less stable.

3. **Velocity/Depth Diversity.** Rated based on the variety of velocity/depth regimes present at a site (slow-shallow, slow-deep, fast-shallow, and fast-deep). As with embeddedness, this metric may result in lower scores in low-gradient streams but will provide a statewide information on the physical habitat found in Maryland streams.

4. **Pool/Glide/Eddy Quality.** Rated based on the variety and spatial complexity of slow- or still-water habitat within the sample segment. It should be noted that even in high-gradient segments, functionally important slow-water habitat may exist in the form of larger eddies. Within a category, higher scores are assigned to segments which have undercut banks, woody debris or other types of cover for fish.

5. **Riffle/Run Quality.** Rated based on the depth, complexity, and functional importance of riffle/run habitat in the segment, with highest scores assigned to segments dominated by deeper riffle/run areas, stable substrates, and a variety of current velocities.

6. **Embeddedness.** Rated as a percentage based on the fraction of surface area of larger particles that is surrounded by fine sediments on the stream bottom. In low gradient streams with substantial natural deposition, the correlation between embeddedness and fishability or ecological health may be weak or non-existent, but this metric is rated in all streams to provide similar information from all sites statewide.

7. **Shading.** Rated based on estimates of the degree and duration of shading at a site during summer, including any effects of shading caused by landforms.

8. **Trash Rating.** The scoring of this metric is based on the amount of human refuse in the stream and along the banks of the sample segment.

Appendix F: Species Names

Common Names of Maryland Fishes

Common Names of Maryland Reptiles and Amphibians

Scientific Names of Maryland Crayfishes

Scientific Names of Maryland Freshwater Bivalves

Common Names of Invasive Plant Species

Names are listed as they should be recorded on MBSS data sheets. Letters in parentheses next to a species name correspond to the state status of the species from the Rare, Threatened, and Endangered Animals of Maryland (April 2010) and from the Maryland Aquatic Species Matrix Team. E = Endangered, T = Threatened, I = In need of Conservation, X = Presumed Extirpated, and N = Nuisance.

Common Names of Maryland Fishes

LAMPREYS:
American brook lamprey (T)
Least brook lamprey
Sea lamprey

HERRINGS:
Alewife
American shad
Blueback herring
Gizzard shad
Hickory shad

CATFISHES:
Blue catfish (N)
Brown bullhead
Channel catfish
Flathead catfish (N)
Margined madtom
Stonecat (E)
Tadpole madtom
White catfish
Yellow bullhead

SUCKERS:
Creek chubsucker
Golden redhorse
Longnose sucker* (X)
Northern hogsucker
Quillback
Shorthead redhorse
White sucker

KILLIFISH:
Banded killifish
Mummichog
Striped killifish

STICKLEBACKS:
Fourspine stickleback
Threespine stickleback

MINNOWS:
Blacknose dace
Bluntnose minnow
Bridle shiner (E)
Central stoneroller
Comely shiner (T)
Common carp
Common shiner
Creek chub
Cutlip minnow
Eastern silvery minnow
Emerald shiner*
Fallfish
Fathead minnow
Golden shiner
Goldfish
Grass carp
Ironcolor shiner (E)
Longnose dace
Mimic shiner (N)*
Pearl dace (T)
River chub
Rosyface shiner
Rosyside dace
Satinfin shiner
Silverjaw minnow
Spotfin shiner
Spottail shiner
Striped shiner (I)
Swallowtail shiner

SCULPINS:
Blue Ridge sculpin
Checkered sculpin
Mottled sculpin
Potomac sculpin

PERCHES:
Banded darter
Chesapeake logperch (T)
Fantail darter
Glassy darter (T)
Greenside darter
Johnny darter
Maryland darter (E)
Rainbow darter
Shield darter
Stripeback darter (E)
Swamp darter (I)
Tessellated darter
Walleye
Yellow perch

SUNFISHES:
Banded sunfish
Black crappie
Blackbanded sunfish (E)
Bluegill
Bluespotted sunfish
Flier (T)
Green sunfish
Largemouth bass
Longear sunfish
Mud sunfish (I)
Pumpkinseed
Redbreast sunfish
Redear sunfish
Rock bass
Smallmouth bass
Warmouth
White crappie

TEMPERATE BASSES:
Striped Bass
White Perch

TROUTS:
Brook trout
Brown trout
Cutthroat trout
Rainbow trout

PIKES:
Chain pickerel
Muskellunge
Northern pike
Redfin pickerel

MISCELLANEOUS:
American eel
Bowfin
Eastern mosquitofish
Eastern mudminnow
Inland silverside
Longnose gar
Northern snakehead (N)
Oriental weatherfish (N)
Pirate perch
Trout-perch (X)

*Historically from and potentially occurring in the Youghiogheny River basin

Common Names of Maryland Reptiles and Amphibians

SALAMANDERS
Allegheny Mountain Dusky Salamander
Common Mudpuppy (X)
Eastern Hellbender (E)
Eastern Mud Salamander
Eastern Red-backed Salamander
Eastern Tiger Salamander (E)
Four-toed Salamander
Green Salamander (E)
Jefferson Salamander
Long-tailed Salamander
Marbled Salamander
Northern Dusky Salamander
Northern Red Salamander
Northern Slimy Salamander
Northern Spring Salamander
Northern Two-lined Salamander
Red-spotted Newt
Seal Salamander
Spotted Salamander
Valley and Ridge Salamander
Wehrle's Salamander (I)

FROGS
American Bullfrog
Barking Treefrog
Carpenter Frog
Cope's Gray Treefrog
Eastern American Toad
Eastern Cricket Frog
Eastern Narrow-mouthed Toad (E)
Eastern Spadefoot
Fowler's Toad
Gray Treefrog
Green Treefrog
Mountain Chorus Frog (E)
New Jersey Chorus Frog
Northern Green Frog
Northern Leopard Frog
Northern Spring Peeper
Pickerel Frog
Southern Leopard Frog
Upland Chorus Frog
Wood Frog

TURTLES
Bog Turtle (T)
Eastern Box Turtle
Eastern Mud Turtle
Eastern Painted Turtle
Eastern River Cooter
Eastern Snapping Turtle
Eastern Spiny Softshell (I)
Midland Painted Turtle
Northern Diamond-backed Terrapin
Northern Map Turtle (E)
Northern Red-bellied Cooter
Red-eared Slider
Spotted Turtle
Stinkpot
Wood Turtle

SNAKES
Coastal Plain Milk Snake
Common Rainbow Snake (E)
Common Ribbonsnake
Eastern Gartersnake
Eastern Hog-nosed Snake
Eastern Kingsnake
Eastern Milk Snake
Eastern Ratsnake
Eastern Smooth Earthsnake
Eastern Wormsnake
Mole Kingsnake
Mountain Earthsnake (E)
Northern Black Racer
Northern Brownsnake
Northern Copperhead
Northern Pinesnake
Northern Red-bellied Snake
Northern Ring-necked Snake
Northern Rough Greensnake
Northern Scarletsnake
Northern Water Snake
Red Cornsnake
Red-bellied Water Snake
Queen Snake
Scarlet Kingsnake
Smooth Greensnake
Southern Copperhead
Southern Ring-necked Snake
Timber Rattlesnake

LIZARDS
Broad-headed Skink
Common Five-lined Skink
Eastern Fence Lizard
Eastern Six-lined Racerunner
Little Brown Skink
Northern Coal Skink (E)
Southeastern Five-lined Skink

Scientific Names of Maryland Crayfishes

Cambarus acuminatus (I)
Cambarus bartonii
Cambarus carinirostris
Cambarus diogenes
Cambarus dubius
Cambarus monongalensis
Cambarus thomaii
Fallicambarus fodiens
Orconectes limosus
Orconectes obscurus
Orconectes rusticus
Orconectes virilis
Procambarus clarkii
Procambarus acutus/zonangulus

Scientific Names of Maryland Freshwater Bivalves

Alasmidonta heterodon (E)
Alasmidonta undulata (E)
Alasmidonta varicosa (E)
Anodonta implicata
Corbicula sp.
Dreissena bugensis
Dreissena polymorpha
Elliptio complanata
Elliptio fisheriana
Elliptio lanceolata
Elliptio producta (I)
Lampsilis cardium
Lampsilis cariosa
Lampsilis radiata
Lasmigona subviridis (E)
Leptodea ochracea
Ligumia nasuta
Pyganodon cataracta
Strophitus undulatus (I)
Utterbackia imbecillis

Common Names of Invasive Plant Species

Autumn Olive
Bamboo sp.
Bull Thistle
Burning Bush
Callery/Bradford Pear
Canada Thistle
Cogon Grass
Daylily (Common)
English Ivy
Garlic Mustard
Giant Hogweed
Japanese Barberry
Japanese Honeysuckle
Japanese Hops
Japanese Knotweed
Japanese Spiraea
Japanese Stiltgrass
Kudzu
Lesser Celandine (Fig Buttercup)
Maiden Grass
Mile-a-Minute
Mimosa
Multiflora Rose
Norway Maple
Oriental Bittersweet
Paulownia (Empress tree)
Phragmites (Common Reed)
Porcelainberry
Privet
Purple Loosestrife
Shrub Honeysuckle
Tree of Heaven
Vinca Vine
Wavyleaf Basketgrass
White Mulberry
Wineberry
Wintercreeper
Wisteria

Appendix G: Chain of Custody Forms

MBSS Water Quality Chain of Custody Sheet

Guidance for MBSS Benthic Macroinvertebrate Sample Chain-of-Custody Sheet

MBSS Benthic Macroinvertebrate Sample Chain-of-Custody Sheet

MBSS Specimen Tracking Data Sheet

MBSS Water Quality Chain of Custody Sheet

UMCES - Appalachian Laboratory
301 Braddock Road
Frostburg, MD 21532

Chain of Custody Record
Maryland Biological Stream Survey
Spring Index Period

Date of Shipment

YYMMDD
_____

Cooler Temperature on Receipt

_____ oC

Analyze For:

1-L Grab: DOC, TDP, TDN, Cl, NO2, NO3, PO4, SO4, NH3, PP, PN
0.5-L Grab: Specific conductance, ANC
Syringe: closed pH

Sample Identification

Site ID	Date YYMMDD	Time (Military)	Site ID	Date YYMMDD	Time (Military)

Field Comments:

Cooler Contents Total Number of:

Syringes _____

1-L Bottles _____

0.5-L Bottles _____

Lab Comments:

Cooler relinquished by: _____
(print name)

_____
(signature)

Cooler received by: _____
(print name)

_____
(signature)

Date and Time of Receipt at Laboratory:

YYMMDD _____ Time (24hr) _____

Page ____ of ____

Guidance for MBSS Benthic Macroinvertebrate Sample Chain-of-Custody Sheet

General

This sheet provides a means of tracking the transfer of benthic macroinvertebrate samples between field collecting crews and DNR field office personnel responsible for processing the samples. If multiple sample containers are delivered for a single site, enter each container on a separate row. If entries are repeated down a row, it is not necessary to enter the information in each cell. Simply use an arrow or quote marks to indicate the information is repeated down the row. Please write as legibly as possible following the guidelines below. The entry of a printed name indicates responsibility of the individual for relinquishing or receiving each sample.

1. Site ID	Enter the site ID just as it appears on the field data form.
2. Collector (print)	Print the name of the person who collected the benthic sample.
3. Collection Date	Enter the date the sample was collected (using DD/MM/YY format) just as it appears on the field data form.
4. Date Delivered to Field Office	Enter the date the sample was delivered to the field office using DD/MM/YY format.
5. Relinquished By (print)	Enter the printed name of the person relinquishing the sample to the appropriate field office staff member.
7. Received By (print)	Enter the printed name of the person receiving the sample at the field office.
8. Field Office Log-In Number	(Done by field office personnel) Enter the Benthic Sample Log-in number.
9. Comments	Place any pertinent comments regarding the delivered samples, including unusual circumstances, here. Examples include "label for sample from site HA-P-056-312 fell off - see label in container" or "some of sample for site HA-P-056-312 spilled while in transport."

If you have questions regarding the use of this sheet or the benthic sample chain-of-custody procedure, call Dan Boward at 410-260-8605.

MBSS Benthic Macroinvertebrate Sample Chain-of-Custody Sheet

Site ID	Number of Buckets	Collector (print)	Collection Date (DD/MM/YY)	Date Delivered to Field Office (DD/MM/YY)	Relinquished by (print)	Received by (print)	Field Office Log Number

Comments _____

_____

_____

Mbss Specimen Tracking Data Sheet

Crew Leader		
Date Delivered MM/DD/YYYY		
Delivered By		
Received By		

Species	**Date Collected** MM/DD/YYYY	**Site ID**	**Voucher Type** Photo or Specimen	**Image no. (s) or No. (s) of specimens**	**Comments**

Appendix H: MBSS Voucher Label

Maryland Biological Stream Survey

SITE ID _____

Cat. No. _____ Family _____

Species _____

Basin _____ Date _____

State _____ County _____

Locality _____

Lat _____ Long _____

Col. By _____

Det. By _____ No. Specimens _____

Appendix I: MBSS Spring Data Sheets

MBSS Spring Index Period Data Sheet

MBSS Spring Faunal Data Sheet

MBSS Vernal Pool Data Sheet

MBSS Spring Habitat Data Sheet

MBSS Spring Facies Map Data Sheet

v. 2014

MBSS SPRING INDEX PERIOD DATA SHEET

Page ☐ of ☐

SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

DATE — Year ☐☐ Month ☐☐ Day ☐☐

CREW: _____

STREAM NAME: _____

LOCALITY: _____

TIME ☐☐☐☐ (Military)

SAMPLEABILITY

☐ Benthos ☐ Facies Mapping

☐ Habitat Assessment

☐ Water Quality

☐ Vernal Pool ☐ Present (Y/N)

Other: _____

SITE ACCESS ROUTE: _____

SAMPLE LABELS

Verified by: _____

QC LABELS

Watershed Code ☐☐☐☐ Segment ☐☐☐ (Letters Only) Type ☐ Year ☐☐☐☐

Dup. (D) or Blank (B): ☐ Verified by: _____

TEMP. LOGGERS

	(Y/N)	(TIME - Military)		
WATER	☐	☐☐☐☐	#	☐☐☐☐☐☐☐☐
AIR	☐	☐☐☐☐	#	☐☐☐☐☐☐☐☐

LOCATION: _____

PHOTODOCUMENTATION

Number		Voucher (Y/N)
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐

BENTHIC HABITAT SAMPLED

☐☐ Riffle

☐☐ Rootwad/Woody Debris

☐☐ Leaf Pack

☐☐ Macrophytes

☐☐ Undercut Banks

Other: _____

SAMPLING CONSIDERATIONS: (NUM. ANODES) _____

STREAM WIDTH (m) ☐☐☐.☐ 0 m ☐☐☐.☐ 75 m

v. 2014

MBSS SPRING FAUNAL DATA SHEET

Page ☐ of ☐

SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐ Reviewer: First _____ / Second _____

HERPETOFAUNA

☐ None Observed

Species	Lifestage: Adult	Larval	Egg	SEEN (Y/N)	HEARD (Y/N)	Number Retained	Number Photos Taken

MUSSELS

☐ None Observed

Species	LIVE	DEAD	Number Retained	Num. Photos Taken

Corbicula LIVE ☐ DEAD ☐ NONE ☐

CRAYFISH

☐ None Observed

Species	Number Retained

Crayfish Burrows (A,P,E) ☐

COMMENTS: _____

v. 2014

MBSS VERNAL POOL DATA SHEET

Page ☐ of ☐

SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐ Reviewer: First _____ / Second _____

Within Transect? (Y/N): ☐ Lat ☐☐.☐☐☐☐☐☐ Long ☐☐.☐☐☐☐☐☐

Vernal Pool ID: V P ☐☐☐

Dimensions: ☐☐☐ m X ☐☐☐ m

Max Depth: ☐☐☐ cm

Landscape Setting: ☐ Upland or Floodplain

Fish Observed? (Y/N): ☐

Anostraca Observed? (Y/N): ☐

Predominant Surrounding Landuse: ☐☐ ☐☐ ☐☐

Distance From Pool →

☐ None Observed

HERPETOFAUNA

Species	Lifestage: Adult	Larva	Egg	Seen (Y/N)	Heard (Y/N)	# Ret.	# Photos Taken
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐

PHOTODOCUMENTATION

Number		Voucher (Y/N)
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐

COMMENTS: _____

Within Transect? (Y/N): ☐ Lat ☐☐.☐☐☐☐☐☐ Long ☐☐.☐☐☐☐☐☐

Vernal Pool ID: V P ☐☐☐

Dimensions: ☐☐☐ m X ☐☐☐ m

Max Depth: ☐☐☐ cm

Landscape Setting: ☐ Upland or Floodplain

Fish Observed? (Y/N): ☐

Anostraca Observed? (Y/N): ☐

Predominant Surrounding Landuse: ☐☐ ☐☐ ☐☐

Distance From Pool →

☐ None Observed

HERPETOFAUNA

Species	Lifestage: Adult	Larva	Egg	Seen (Y/N)	Heard (Y/N)	# Ret.	# Photos Taken
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐
_____	☐	☐	☐	☐	☐	☐	☐

PHOTODOCUMENTATION

Number		Voucher (Y/N)
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐

COMMENTS: _____

v. 2014

MBSS SPRING HABITAT DATA SHEET

Page ☐ of ☐

SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐ Reviewer: First _____ / Second _____

DATE — Year ☐☐ Month ☐☐ Day ☐☐

☐☐☐☐ Dist. from Nearest Road to Site (m)

☐☐ Trash Rating 0 - 20

LANDUSE (Y/N)

- ☐ Old Field
- ☐ Deciduous Forest
- ☐ Coniferous Forest
- ☐ Wetland
- ☐ Surface Mine
- ☐ Landfill
- ☐ Residential
- ☐ Commercial/Industrial
- ☐ Cropland
- ☐ Pasture
- ☐ Orchard/Vineyard/Nursery
- ☐ Golf Course

RIPARIAN VEGETATION
(facing upstream)

	LEFT BANK	RIGHT BANK
Width (50m max)	☐☐	☐☐
Adj. Land Cover	☐☐	☐☐
Veg Type	☐☐☐☐	☐☐☐☐
Buffer Breaks (Y/N)	☐	☐

ROAD CULVERT

- Present in Segment? (Y/N) ☐
- Sampleable? (Y/N) ☐
- Width of Culvert (m) ☐☐
- Length of Culvert (m) ☐☐

STREAM GRADIENT

	Location (m)	Height (m)
1	☐☐	☐.☐☐
	☐☐	☐.☐☐
2	☐☐	☐.☐☐
	☐☐	☐.☐☐
3	☐☐	☐.☐☐
	☐☐	☐.☐☐

BUFFER BREAKS

	LEFT BANK	RIGHT BANK
Storm Drain	☐	☐
Tile Drain	☐	☐
Imperv. Drainage	☐	☐
New Construction	☐	☐
Orchard	☐	☐
Crop	☐	☐
Pasture	☐	☐
Gully	☐	☐
Dirt Road	☐	☐
Gravel Road	☐	☐
Raw Sewage	☐	☐
Railroad	☐	☐

Buffer Break Types (M = Minor; S = Severe)

CHANNELIZATION

☐ Evidence of Channel Straightening or Dredging (Y/N)

TYPE	EXTENT (m) LEFT BANK	BOTTOM	RIGHT BANK
Concrete	☐☐	☐☐	☐☐
Gabion	☐☐	☐☐	☐☐
Rip-Rap	☐☐	☐☐	☐☐
Earthen Berm	☐☐	☒☒	☐☐
Dredge Spoil Off Channel	☐☐	☒☒	☐☐
Pipe Culvert	☐☐	☐☐	☐☐

Actual Site Midpoint Coordinates
(Taken at Time of Sampling)

Lat ☐☐.☐☐☐☐☐

Long ☐☐.☐☐☐☐☐

Stream Blockages

Stream Block Ht. (m) ☐.☐☐

Stream Block Type ☐☐

Lat ☐☐.☐☐☐☐☐

Long ☐☐.☐☐☐☐☐

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MBSS SPRING FACIES MAP DATA SHEET

Page ☐ of ☐

SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

DATE Year ☐☐ Month ☐☐ Day ☐☐

	L	Center	R
75m	Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐		Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐
50m	Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐		Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐
25m	Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐		Dominant Substrate ☐ Subdominant Substrate ☐ Depth ☐ Velocity ☐
0m			

Classifications for **Dominant** and **Subdominant Substrate** Categories

Y = Silt/Clay (< .062mm)
S = Sand (.062 - 2mm)
G = Gravel (2 - 64mm)
C = Cobble (64 - 256mm)
B = Boulder (256-4096mm)
K = Bedrock (> 4096mm)

Classifications for Average Stream **Depth** Categories

1 = Shallow (< 0.5 m)
2 = Moderately Deep (0.5 m - 1.0 m)
3 = Deep (> 1.0 m)

Classifications for Average Stream **Velocity** Categories

1 = Slow (0-0.3 m/s)
2 = Fast (> 0.3 m/s)

COMMENTS

Appendix J: MBSS Summer Data Sheets

MBSS Summer Index Data Sheet

MBSS Summer Habitat Data Sheet

MBSS Summer Fauna Data Sheet

MBSS Stream Salamander Data Sheet

MBSS Fish Data Sheet

MBSS Game Fish Length Data Sheet

MBSS Photo Data Sheet

MBSS Round 1 Repeat Data Sheet

Crib Sheet

v. 2014

MBSS SUMMER INDEX DATA SHEET

Page ☐ of ☐

SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

DATE — Year ☐☐ Month ☐☐ Day ☐☐

CREW: _____

TIME ☐☐☐☐ (Military)

COMMENTS: _____

SAMPLEABILITY

S = Sampleable
1 = Dry Streambed
2 = Too Deep
3 = Marsh, no defined channel
4 = Excessive Riparian Vegetation
5 = Impoundment
6 = Tidally Influenced
7 = Landowner Permission Denied
8 = Unsafe (Describe in Comments)
9 = Beaver
10 = Other: _____

- ☐ Electrofishing
- ☐ Habitat
- ☐ Water Quality
- ☐ Herpetofauna
- ☐ Salamanders
- ☐ Crayfishes
- ☐ Mussels
- ☐ Aquatic Plants
- ☐ Exotic Plants
- ☐ Geomorphology

EXOTIC PLANTS

☐ None Observed

Species	Relative Abundance (P or E)
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐
_____	☐

TEMPERATURE LOGGERS

Air Logger Present? (Y/N) ☐

Water Logger De-watered? (Y/N) ☐

AQUATIC VEGETATION

(A,P, or E)

- Submerged Aquatic Vegetation ☐
- Emergent Aquatic Vegetation ☐
- Floating Aquatic Vegetation ☐

DIDYMO

(A,P, or E) ☐ Voucher (Y/N) ☐

PHOTODOCUMENTATION

Number		Voucher (Y/N)
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐
☐☐☐☐	_____	☐

COMMENTS: _____

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MBSS SUMMER HABITAT DATA SHEET

Page ☐ of ☐

SITE | Watershed Code ☐☐☐☐ | Segment ☐☐☐ | Type ☐ | Year ☐☐☐☐ | Reviewer: First _____ / Second _____

BANK EROSION

	Left Bank	Right Bank
Extent (m)	☐☐	☐☐
Severity ___	☐	☐
Average Height (m)	☐☐☐	☐☐☐

Severity
0 = none
1 = min
2 = mod
3 = severe

BAR FORMATION & SUBSTRATE

Severity ___
0 = none
1 = min
2 = mod
3 = severe

☐ Cobble
☐ Gravel
☐ Sand
☐ Silt/Clay

HABITAT ASSESSMENT

1. Instream Habitat (0-20) ☐☐
2. Epifaunal Substrate (0-20) ☐☐
3. Velocity/Depth Diversity (0-20) ☐☐
4. Pool/Glide/Eddy Quality (0-20) ☐☐
   Extent (m) ☐☐
5. Riffle/Run Quality (0-20) ☐☐
   Extent (m) ☐☐
6. Embeddedness (%) ☐☐☐
7. Shading (%) ☐☐☐

FLOW

Lat. Loc. (m)	Depth (cm)	Velocity (m/s)
0 0 0 0	0 0 0	0 0 0
☐☐☐☐	☐☐☐	☐☐☐

STREAM CHARACTER

☐ Braided	☐ Gravel	☐ Boulder >2m
☐ Riffle	☐ Sand	☐ Boulder <2m
☐ Run/Glide	☐ Silt/Clay	☐ Beaver Pond
☐ Deep Pool (>= 0.5m)	☐ Cobble	☐ Overhead Cover
☐ Shallow Pool (< 0.5m)	☐ Bedrock	☐ Undercut Bank
		☐ Orange Floc

A = Absent P = Present E = Extensive

Woody Debris

☐☐ No. of Instream Woody Debris
☐☐ No. of Dewatered Woody Debris
☐☐ No. of Instream Rootwads
☐☐ No. of Dewatered Rootwads

Maximum Depth (cm) ☐☐☐

	Wetted Width (m)	Thalweg Depth (cm)	Thalweg Velocity (m/s)
0 m	☐☐☐	☐☐☐	☐☐☐
25 m	☐☐☐	☐☐☐	☐☐☐
50 m	☐☐☐	☐☐☐	☐☐☐
75 m	☐☐☐	☐☐☐	☐☐☐

Alternative Flow Measurements

Distance (cm) ☐☐☐
Depth (cm) ☐
Width (cm) ☐☐
Time (sec) 1. ☐☐☐
2. ☐☐☐
3. ☐☐☐

COMMENTS: _____

v. 2014

MBSS SUMMER FAUNA DATA SHEET

SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First ______ / Second ______

STREAM SALAMANDERS

☐ None Observed

Species	Electrofishing Catch Adult	Electrofishing Catch Larva	Transect Catch Adult	Transect Catch Larva	Number Retained	Number Photos Taken

OTHER HERPETOFAUNA

☐ None Observed

Species	Lifestage Adult	Lifestage Larva	Lifestage Egg	Number Retained	Num. Photos Taken

MUSSELS

☐ None Observed

Species	Live	Dead	Number Retained	Num. Photos Taken

Corbicula — LIVE ☐ DEAD ☐ NONE ☐

CRAYFISH

☐ None Observed

Crayfish Burrows (Absent, Present, Extensive) ☐

Species	Incidental Catch? (Y/N)	1st Pass Catch (Total)	2nd Pass Catch (Total)	Number Retained

COMMENTS: ______________________________

v. 2014

MBSS STREAM SALAMANDER DATA SHEET

Page ☐ of ☐

SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First ____ / Second ____

DATE — Year ☐☐ Month ☐☐ Day ☐☐

Seeps Present? (Y/N) ☐

Habitat Composition (%)

☐☐☐ Stream Corridor

☐☐☐ Seeps

	Stream Corridor	Seeps
All Available Habitat Sampled? (Y/N)	☐	☐
Time Searched (Max 60 min)	Minutes ☐☐ : Seconds ☐☐	Minutes ☐☐ : Seconds ☐☐
Available Habitat Left Unsampled (%)	☐☐	☐☐

COMMENTS

____

____

____

____

____

____

____

____

____

____

☐ None Observed

SPECIES	ADULTS Abundance Stream Corridor	ADULTS Abundance Seeps	ADULTS Num. Photos Taken	LARVAE Abundance Stream Corridor	LARVAE Abundance Seeps	LARVAE Num. Photos Taken

v. 2014

MBSS FISH DATA SHEET

Page ____ of ____

SITE Watershed Code ____ Segment ____ Type ____ Year ____

Reviewer: First _____ / Second _____

	(Y/N)
Fish Move. During Net Installation?	
Bottom Visible in all Areas of Seg.?	
Same Water Clarity - 2nd Pass?	
Length of Seg. Sampled (m)	
Fish Captured?	
Gamefish?	

	Unit ____	Unit ____	Unit ____	Unit ____	Unit ____	Unit ____
Anodes/Unit						
Begin 1st P.						
Begin 2nd P.						
End						
Voltage						

SPECIES	1st Pass Catch (Total)	2nd Pass Catch (Total)	Anomalies (Y/N)	Number Retained	Number of Photos	Comments

Aggregate Fish Biomass ____ (g)

v. 2014

MBSS GAME FISH LENGTH DATA SHEET

Page ☐ of ☐

SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

SPECIES	LENGTH (TL; mm)	SPECIES	LENGTH (TL; mm)

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MBSS PHOTO DATA SHEET

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SITE — Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

Number	PHOTODOCUMENTATION	Voucher (Y/N)

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MBSS ROUND 1 REPEAT DATA SHEET

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SITE Watershed Code ____ Segment ____ Type ____ Year ____

Reviewer: First _____ / Second _____

Habitat Assessment

Channel Alteration (0-20) .....

Bank Stability (0-20) .....

Remoteness (0-20) .....

Minimum Buffer Zone Width (m) .....

Buffer Type .....

Adjacent Land Cover .....

Fish Anomalies

Species w/ Anomalies	Number of Affected Individuals (max. 100 per species)																	
	Anomaly Type			Anomaly Type			Anomaly Type			Anomaly Type			Anomaly Type			Anomaly Type		

Anomaly Types

Body Surfaces and Fins
- AN = Swelling of the Anus
- AW = Anchor Worm
- BL = Black Spot
- BS = Body Shape
- CL = Fin Cloudiness
- CT = Cut
- DI = Discoloration
- DM = Deformities of the Mandible
- DV = Deformities of the Vertebral Column
- EP = Visible External Parasites
- FD = Fin Deformed or Missing
- FI = Fin Erosion
- FU = Fungus
- GR = Growths/Cysts
- HK = Hooking Injury
- HM = Hemorrhaging
- IK = Ich
- LE = Leeches
- OT = Other (define in comments section)
- RE = Red Spot
- RS = Raised Scales
- SC = Scale Deformities
- UL = Ulcerations/Lesions

Eyes
- CA = Cataract
- EC = Eye Cloudiness
- EH = Eye Hemorrhage
- NO = Eye missing
- OR = Depression into the Orbits
- PO = Exopthalmia (pop eye)

Comments

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CRIB SHEET

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	Watershed Code	Segment	Type	Year
SITE				

_____ PASS			_____ PASS		
Species	Tally	Anomalies	Species	Tally	Anomalies

Appendix K: MBSS Geomorphology Data Sheets

MBSS Geomorphology Cross-Section Data Sheet

MBSS Flood-Prone Area Data Sheet

MBSS Cross-Section Pebble Counts Data Sheet

MBSS GEOMORPHOLOGY CROSS-SECTION DATA SHEET

SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

Cross-Section Within Site? (Y/N) ☐

Cross-Section Within Riffle? (Y/N) ☐
(riffle may not be present on Coastal Plain)

	Distance, Point or	Back-Sight	Fore-Sight	COMMENTS:
	STATION	B S	F S	
Item	ft	ft	ft	
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
11				
12				
13				
14				
15				
16				
17				
18				
19				
20				
21				
22				
23				
24				
25				
26				
27				
28				

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MBSS FLOOD-PRONE AREA DATA SHEET

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SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

1.)

(A) ☐☐.☐☐ ft

– (B) ☐☐.☐☐ ft

(C) ☐☐.☐☐ ft

A = Elevation at average water depth (from Cross-Section Data Sheet)

B = Mean bankfull depth (from regional curve)

C = Bankfull elevation (A-B)

D = Thalweg elevation (from Cross-Section Data Sheet)

E = Max bankfull depth (D-C)

F = 2x max bankfull depth (E x 2)

G = Flood-prone area elevation (D-F)

2.)

(D) ☐☐.☐☐ ft

– (C) ☐☐.☐☐ ft

(E) ☐☐.☐☐ ft

3.)

X 2 = (F) ☐☐.☐☐ ft

4.)

(D) ☐☐.☐☐ ft

– (F) ☐☐.☐☐ ft

(G) ☐☐.☐☐ ft ← FLOOD-PRONE ELEVATION

Flood-Prone Elevation >50m from edge of water? (Y/N) ☐

FLOOD-PRONE AREA WIDTH ☐☐☐.☐ ft

Comments/Notes:

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MBSS CROSS-SECTION PEBBLE COUNTS DATA SHEET

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SITE Watershed Code ☐☐☐☐ Segment ☐☐☐ Type ☐ Year ☐☐☐☐

Reviewer: First _____ / Second _____

		Range (mm)	Particle Tally	Total
SILT/CLAY	Silt/Clay	<.062		
SAND	Very Fine	.062 - .125		
	Fine	.125 - .25		
	Medium	.25 - .50		
	Coarse	.50 - 1.0		
	Very Coarse	1.0 - 2		
GRAVEL	Very Fine	2 - 4		
	Fine	4 - 6		
	Fine	6 - 8		
	Medium	8 - 11		
	Medium	11 - 16		
	Coarse	16 - 22		
	Coarse	22 - 32		
	Very Coarse	32 - 45		
	Very Coarse	45 - 64		
COBBLE	Small	64 - 90		
	Small	90 - 128		
	Large	128 - 180		
	Large	180 - 256		
BOULDER	Small	256 - 362		
	Small	362 - 512		
	Medium	512 - 1024		
	Large	1024 - 2048		
	Very Large	2048 - 4096		
BEDROCK	Bedrock	>4096		
			Grand Total	