United States Environmental Protection Agency Office of Water
Office of Environmental Information Washington, DC
EPA *841-B-07-007*

## National Rivers and Streams Assessment

# Quality Assurance Project Plan

# Rivers and Streams Assessment

**Final Document December 2010**

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National Rivers and Streams Assessment (NRSA)
Quality Assurance (QA) Project Plan

Management Approvals

Signature indicates that this QAPP is approved and will be implemented in conducting this project.

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# QUALITY ASSURANCE PROJECT PLAN REVIEW & DISTRIBUTION ACKNOWLEDGMENT AND COMMITMENT TO IMPLEMENT for National Rivers and Streams Assessment

We have read the QAPP and the methods manuals for the National Rivers and Streams Assessment listed below. Our agency/organization agrees to abide by its requirements for work performed under our cooperative agreement for Demonstration of Randomized Design for Assessment of National Rivers and Streams (under CWA 104(b) (3)).

| | |
|---|---|
| *Quality Assurance Project Plan* | *EPA-841-B-07-007* |
| *Site Evaluation Guidelines* | *EPA-841-B-07-008* |
| *Field Operations Manual* | *EPA-841-B-07-009* |
| *Laboratory Methods Manual* | EPA-841-B-07-010 |

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Please return the signed original to the EPA QA officer for this cooperative agreement:

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Retain a copy for your files.

## NOTICE

The complete documentation of overall NRSA project management, design, methods, and standards is contained in four companion documents, including:

*National Rivers and Streams Assessment: Quality Assurance Project Plan EPA-841-B-07-007*
*National Rivers and Streams Assessment: Site Evaluation Guidelines EPA-841-B-07-008*
*National Rivers and Streams Assessment: Field Operations Manual EPA-841-B-07-009*
*National Rivers and Streams Assessment: Laboratory Methods Manual EPA 841-B-07-010*

This document (*Quality Assurance Project Plan*) contains elements of the overall project management, data quality objectives, measurement and data acquisition, and information management for the NRSA, and is based on the guidelines developed and followed in the Western Environmental Monitoring and Assessment Program (Peck et al. 2003). Methods described in this document are to be used specifically in work relating to the NRSA. All Project Cooperators must follow these guidelines. Mention of trade names or commercial products in this document does not constitute endorsement or recommendation for use. More details on specific methods for site evaluation, field sampling, and laboratory processing can be found in the appropriate companion document(s) listed above.

The suggested citation for this document is:

USEPA. 2008 (draft). National Rivers and Streams Assessment: Integrated Quality Assurance Project Plan. EPA/841/B-07/007. U.S. Environmental Protection Agency, Office of Water and Office of Research and Development, Washington, DC.

## DISTRIBUTION LIST

This QA Project Plan and associated manuals or guidelines will be distributed to the following EPA, Tetra Tech, Inc. (Tt), and Great Lakes Environmental Center (GLEC) senior staff participating in the NRSA and to State Water Quality Agencies or cooperators who will perform the field sampling operations. The Tt and GLEC QA Officers will distribute the QA Project Plan and associated documents to participating project staff at their respective facilities and to the project contacts at participating laboratories, as they are determined.

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# 1.0 PROJECT PLANNING AND MANAGEMENT

## 1.1 Introduction

Several recent reports have identified the need for improved water quality monitoring and analysis at multiple scales. In 2000, the General Accounting Office (USGAO, 2000) reported that EPA and states cannot make statistically valid inferences about water quality (via 305[b] reporting) and lack data to support key management decisions. In 2001, the National Research Council (NRC, 2000) recommended EPA and states promote a uniform, consistent approach to ambient monitoring and data collection to support core water quality programs. In 2002, the H. John Heinz III Center for Science, Economics, and the Environment (Heinz Center, 2002) found there are inadequate data for national reporting on fresh water, coastal and ocean water quality indicators. The National Association of Public Administrators (NAPA, 2002) stated that improved water quality monitoring is necessary to help states make more effective use of limited resources. EPA's Report on the Environment 2003 (USEPA, 2003) says that there is insufficient information to provide a national answer, with confidence and scientific credibility, to the question, "What is the condition of U.S. waters and watersheds?"

In response to this need, the U.S. Environmental Protection Agency (EPA) Office of Water (OW), in concert with EPA's Office of Research and Development (ORD) and the 10 EPA Regions, conceived of the National Aquatic Resource Surveys (NARS), which includes the National Rivers and Streams Assessment (NRSA) - a national assessment of the condition of rivers and streams in the conterminous U.S. NRSA is the first assessment on flowing waters to be based on data collected using the same field and laboratory protocols and based on a statistical survey design that would allow inferences about all waters based on a sample of the rivers and streams across the country. The desire is to implement this effort in cooperation with the States and other entities eligible for 106 funding. NRSA builds upon the Environmental Monitoring and Assessment Program's (EMAP) Western Study implemented by ORD, the EPA Regions, States and Tribal nations in 12 western states and the Wadeable Streams Assessment (WSA) undertaken in 2004. NRSA will provide the baseline for rivers and streams across the country and regionally across many indicator types, as well as a comparison of stream information to the original WSA.

The NRSA Quality Assurance Project Plan (QAPP) is designed to support the participants in this project and to ensure that the final assessment is based on high quality data and information. The QAPP contains elements of the overall project management, data quality objectives, measurement and data acquisition, and information management for the NRSA. The participants in the NRSA have agreed to follow this QAPP and the protocols and design laid out in this document.

The NRSA is designed to answer key questions asked by Congress, the public, and decision makers, such as:

- What's the extent of waters that support healthy ecosystems, recreation, and fish consumption?

- How widespread are the most significant water quality problems?
- Over time and as additional surveys are implemented, these data will also contribute to answer questions such as:
- Is water quality improving?
- Are we investing in restoration and protection wisely?

Ecological assessments via the NRSA will provide estimates (with quantifiable uncertainty) of the biological integrity of macroinvertebrate, fish, phytoplankton and periphyton communities in streams and rivers. Recreational indicators such as fecal contaminants and fish tissue will be collected to look at human health related issues. Additionally, indicators of physical habitat condition such as bank stability, channel alterations, and invasive species; basic water chemistry; and watershed characteristics will also be collected to assist in explaining the patterns found in biological communities across the country.

### 1.2 NRSA Project Organization

The major areas of activity and responsibilities are described here and illustrated in Figure 1. The overall coordination of the project will be provided by EPA's Office of Water (OW) in Washington, DC, with technical support from the Western Ecology Division (WED) of the Office of Research and Development (ORD) in Corvallis, Oregon and the ten EPA Regional Offices. This comprehensive quality assurance (QA) program has been established to ensure data integrity and provide support for the reliable interpretation of the findings from this project.

Program level QA will be the responsibility of the OWOW QA Officer and the Project QA Officer. A QA records system will be used to maintain indefinitely a permanent hardcopy file of all NRSA documentation from site selection to data analysis. This will be housed in OW Headquarters Office.

The primary responsibilities of the principals and cooperators are as follows:

***Project Management***:

*EPA Project Leader* – provides overall coordination of the project and makes decisions regarding the proper functioning of all aspects of the project. Makes assignments and delegates authority, as needed to other parts of the project organization.

*EPA Project QA Lead* - provides leadership, development and oversight of project level quality assurance for NRSA in Office of Water

*EPA ORD Technical Advisor* – advises the Project Leader on the relevant experiences and technology developed within ORD's EMAP that are to be used in this project. Serves as primary point-of-contact for project coordination in the absence or unavailability of Project Leader.

*Project Coordination* - contractor providing day-to-day coordination of field implementation as well as technical development of analysis of data.

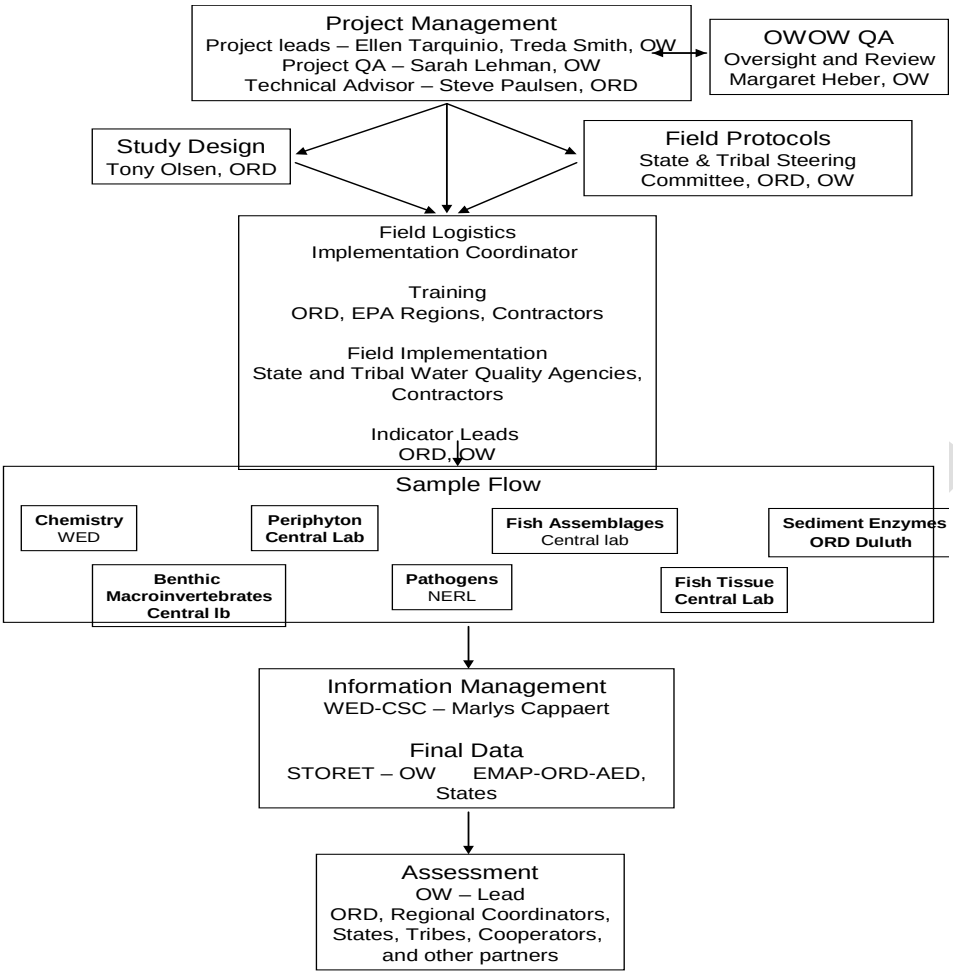


**Figure 1.** NRSA Project Organization

***Study Design**:*

**Objectives: The study is designed to sample 1800 probabilistic, 200 repeat sites and 200 reference sites (2200 total) river and stream sites across the country.**

The objectives, or design requirements, for the National Rivers and Streams Assessment are to produce:

1. Estimates of the 2008-2009 status of flowing waters nationally and regionally (9 aggregated Omernik ecoregions),
2. Estimates of the 2008-2009 status of wadeable streams and non-wadeable rivers nationally and regionally (9 aggregated Omernik ecoregions),
3. Estimates of the 2008-2009 status of urban flowing waters nationally,
4. Estimates of the change in status in wadeable streams between 2008-2009 and 2004, nationally and regionally (9 aggregated Omernik ecoregions).

**Target population:** The target populations consists of all streams and rivers within the 48 contiguous states that have flowing water during the study index period excluding portions of tidal rivers up to head of salt defined as .05 ppt measured in the field). The study index period extends from May to October and is characterized by low flow or base flow conditions. The target population includes the Great Rivers (i.e. main stem of the Mississippi River). Run-of-the-river ponds and pools are included while reservoirs are excluded (those that have greater than 7 day retention period).

**Sample Frame:** The sample frame was derived from the National Hydrography Dataset (NHD), in particular NHD-Plus. Attributes from NHD-Plus and additional attributes added to the sample frame that are used in the survey design include: (1) state, (2) EPA Region, (3) NAWQA Mega Region, (4) Omernik Ecoregion Level 3 (NACEC version), (4) WSA aggregated ecoregions (nine and three regions), (5) Strahler order, (6) Strahler order categories ($1^{st}$, $2^{nd}$, …, $7^{th}$ and $8^{th}$ +), (6) FCode, (7) Urban, and (8) Frame07.

**Expected sample size:** Expected sample size is 1800 flowing water sites: 450 sites revisited from the WSA, 450 new sites from $1^{st}$ to $4^{th}$ order, and 900 new sites from $5^{th}$ to $10^{th}$ order.

**Over sample:** No over sample sites were selected for the WSA_Revisit design. The expectation is that all, or almost all, of the 450 sites selected will be sampled given they were sampled previously. For the NRSA design, the over sample is nine times the expected sample size within each state. The large over sample size was done to accommodate those states who may want to increase the number of sites sampled within their state for a state-level design.

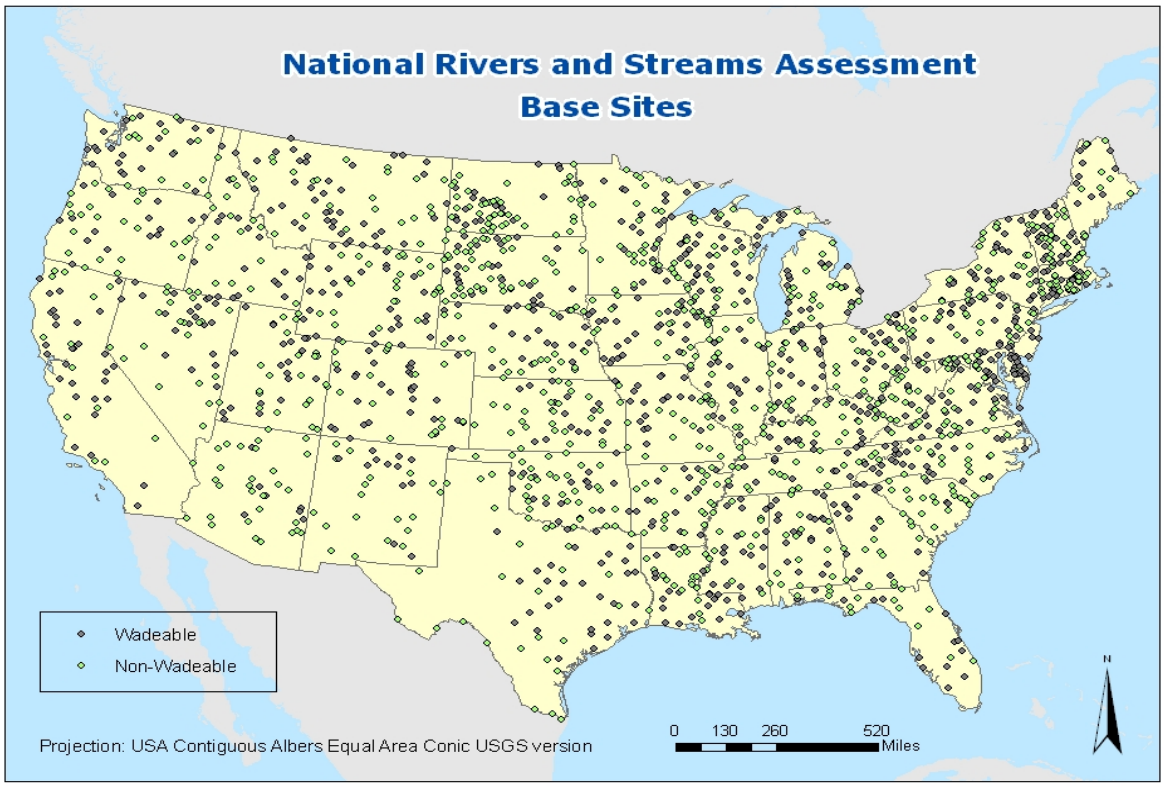


**Figure 2.** NRSA Base Sites

***Field Protocol Development****:* The field sampling protocols are based on protocols developed by ORD for use in the EMAP program and were developed with the purpose of providing consistent and representative information across the country. During the initial design phase of the project, collaborators and partners worked to refine those protocls for use in the NRSA. This involved modifications to the original protocols used in the EMAP program for use in the Great Rivers, tidal systems, and sites that were in between a wadeable and a boatable system. New advance in the field, such as the incorporation of surveyors levels for a more accurate measure of slope in wadeable sites were also incorporated based on the consensus of the partners indicator workgroups. In addition, OWOW directed development of fecal bacteria (*Enterococci*) indicator sampling protocols and OST developed field protocols for the fish tissue indicator.

***Field Logistics Coordinator***– a contractor who functions on behalf of the Project Leader to support all phases of the field implementation of the project. Primary responsibility is to ensure all aspects of the project, i.e., technical, logistical, organizational, are operating as smoothly as possible. Serves as point-of-contact for questions from field crews and cooperators for all activities.

***Training*** - Ten training sessions will be conducted in various locations throughout the US per field year ( ten in 2008 and ten in 2009). An initial training session focusing on training the trainers was held in March 2008 and in March 2009. Headquarters, GLEC/Tetra Tech (contract), and participants from the train the trainers session conducted the remaining training sessions. When possible, a monitoring specialist

from each EPA Regional Office also participated in each of the trainings. Each field crew must have a crew leader who has received 3 days of lecture and field training to prepare them for this study. They must also have a fish technical lead who has participated in the training and received prior approval from the EPA Project Lead. At the end of the training period, each team will conduct a day long sampling on their own under the watch of the trainers. This field readiness review will be the final QA check of the training sessions. Additionally, all field crews will be audited early in their sampling schedule to be certain any corrections will be made at the onset of sampling.

***Field Implementation*** - States, Tribes, Interstate Agencies, and contract crews will conduct the field implementation to collect samples using the NRSA protocols.

***Field Quality Evaluation and Assistance Reviews (auditing)*** - Each field team will be visited by a trained team from either an EPA Region, Headquarters, GLEC, or Tetra Tech. The purpose of this field evaluation and assistance review is to observe the crews implementing the protocols as trained and provide any assistance or corrections necessary. This is intended to catch deviations from the protocols before they become widespread.

***Sample Flow***: Field samples will be shipped by the crews to one of several locations. All water samples will be sent to the Western Ecology Division laboratory staffed by Dynamac. All biological samples will be sent to a national contract lab for analysis or the prior approved state biological laboratory. Enterococci samples will be sent to Region 1 Lab staffed by Tech Law for analysis. The fish tissue samples will be sent to GLEC for homogenization and filleting. The field data sheets will be shipped to the Western Ecology Division information management team staffed by CSC for scanning and entry into the database. Each of the organizations processing samples will electronically transfer the results to CSC using the naming conventions and standards provided by CSC.

***Information Management:*** The first stage of data processing will be to take the input from each of the responsible laboratories and enter them into a common database for final verification and validation. Once the final data sets are made available for the assessment, copies of the data will be transferred to EPA's STORET and EPA's EMAP dataset for long-term storage and access. Working copies of the final data sets will be distributed to the States and Cooperators and maintained at WED for analysis leading to the assessment.

***Assessment:*** The final assessment will be developed by a team, led by OW, that will include Office of Water, Office of Environmental Information, several ORD research facilities, EPA Regional Monitoring Coordinators, interested States/Tribes, and Cooperators. All States/Tribes will be invited to participate in a collaborative process to interpret results and shape the data assessment and report. The final assessment will include an appendix describing the quality of the data used in the assessment.

### *1.2.1 Project Schedule*

The U.S. EPA has responded to a State and OW goal to report on the quality of the Nation's

rivers and streams by no later than December, 2011. Tasks leading up to the final report are described throughout the QAPP.

## 1.3 Scope of QA Project Plan

This QA Project Plan addresses all aspects of the data acquisition efforts of the NRSA, which focuses on the 2008 and 2009 sampling of 2200 river and stream sites in the contiguous United States. This QA plan also deals with the data integration necessary between the WSA, NRSA, and EMAP Western Pilot Study (2001-2004) to create one complete report on the ecological status of the Nation's rivers and streams.

Relevant Companion documents to this QAPP are: NRSA: Site Evaluation Guidelines, NRSA: Field Operations Manual, and NRSA: Laboratory Methods Manual ( See introductory pages for citation information for each document).

### *1.3.1 Overview of Field Operations*

Field data acquisition activities are implemented for the NRSA (Table 1-1), based on guidance developed for earlier EMAP studies (Baker and Merritt 1990). Survey preparation is initiated with selection of the sampling locations by the EMAP Design group (WED in Corvallis). The list of sampling locations is distributed to the EPA Regional Monitoring Coordinators and all cooperators. With the sampling location list, Cooperator's field crews can begin site reconnaissance on the primary sites and alternate replacement sites and begin work on obtaining access permission to each site. Specific procedures for evaluating each sampling location and for replacing non target sites are documented in the *NRSA: Site Evaluation Guidelines*. Scientific collecting permits from State and Federal agencies will be procured, as needed by the respective State or cooperating organization. The field teams will use standard field equipment and supplies which are being provided by EPA and GLEC. Field logistic coordinators (GLEC and Tetra Tech) will work with Regional Monitoring Coordinators, Cooperators, States, and Contractors to make certain the field crews have the equipment and supplies they require in a timely fashion. Detailed lists of equipment required for each field protocol, as well as guidance on equipment inspection and maintenance, are contained in the Field Operations Manual.

**Table 1-1.** Critical logistics elements (from Baker and Merritt, 1990)

| Logistics Plan Component | Required Elements |
|---|---|
| Project Management | Overview of Logistic Activities<br>Staffing and Personnel Requirements<br>Communications |
| Access and Scheduling | Sampling Schedule<br>Site Access<br>Reconnaissance |
| Safety | Safety Plan<br>Waste Disposal Plan |
| Procurement and Inventory Control | Equipment, Supplies, and Services Requirements<br>Procurement Methods and Scheduling |
| Training and Data Collection | Training Program<br>Field Operations Scenario<br>Laboratory Operations Scenarios<br>Quality Assurance<br>Information Management |
| Assessment of Operations | Field Crew Debriefings<br>Logistics Review and Recommendations |

Field measurements and samples are collected by trained teams. Each Crew Leader will be trained at an EPA-sponsored training session prior to the start of the field season along with as many crew members as possible. Half of the field team musthave participated in an official NRSA training. Fish leads must also attend the training, as well as receive prior approval by EPA Project Lead to serve in this role. Field quality evaluation and assistance review visits will be completed for each team. Typically, each team is comprised of 4-5 members. The number and size of teams depends on the duration of the sampling window, geographic distribution of sampling locations, number and complexity of samples and field measurements, and other factors. The training program stresses hands-on practice of methods, comparability among crews, collection of high quality data and samples, and safety. Training will be provided in ten central locations for cooperators and contractors each year. Project organizations responsible for training oversight are identified in Figure 1. Training documentation will be maintained by the EPA HQ, Tetra Tech and GLEC Training Support Team.

For each sampling location, a dossier will be prepared by the field crew and contains the following applicable information: road maps, copies of written access permissions, scientific collection permits, coordinates of index sites, information brochures on the program for interested land owners, a topographic map with the index site location marked, and local area emergency numbers. Team leaders will contact landowners at least 2 days before the planned sampling date. As the design requires repeat visits to selected sampling locations, it is important for the field teams to do everything possible to maintain good relationships with landowners. This includes prior contacts, respect of special requests, closing gates, minimal site disturbance, and removal of all materials including flagging and trash.

A variety of methods may be used to access a site, including vehicles and boats. Some

sampling locations require teams to hike in, transporting all equipment in backpacks. For this reason, ruggedness and weight are important considerations in the selection of equipment and instrumentation. Teams may need to camp out at the sampling location and if this is the case Teams must be equipped with the necessary camping equipment.

The site verification process is shown in Figure 3. Upon arrival at a site, the location is verified by a Global Positioning System (GPS) receiver, landmark references, and/or local residents. Samples and measurements for various indicators are collected in a specified order (Figure 4). This order has been set up to minimize the impact of sampling for one indicator upon subsequent indicators; for example, water chemistry samples from rivers and streams are collected before collecting benthic invertebrates as the benthic invertebrate method calls for kicking up sediments. All methods are fully documented in step-by-step procedures in the *NRSA: Field Operations Manual* (USEPA 2008). The manual also contains detailed instructions for completing documentation, labeling samples, any field processing requirements, and sample storage and shipping. Any revision of methods must be approved in advance by the EPA Project Leader. Field communications will be available through Field Coordinators, regularly scheduled conference calls, a Communications Center, or an electronic distribution.

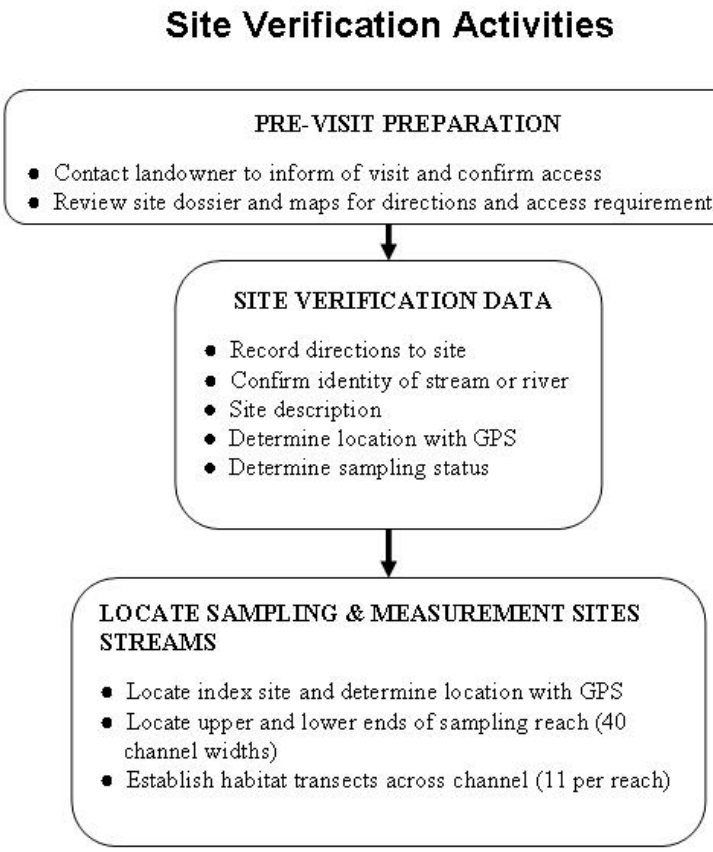


**Figure 3.** Site verification activities for river and stream field surveys.

Standardized field data forms are provided to the field crews as the primary means of data recording. On completion, the data forms are reviewed by a field crew member other than the person who initially entered the information. Prior to departure from the field site, the field team leader reviews all forms and labels for completeness and legibility and ensures that all samples are properly labeled and packed. Each site has a unique identifier (Site ID) provided by the design. All jars from a site have a predetermined sample number that is preprinted on the labels provided to the field crews. If additional jars are needed, extra labels are provided.

On return from a field sampling site (either to the field team's home office or to a motel), completed data forms are sent to the information management staff at WED for entry into a computerized data base. At WED, electronic data files are reviewed independently to verify that values are consistent with those recorded on the field data form or original field data file.

Samples are stored or packaged for shipment in accordance with instructions contained in the field manual. Samples which must be shipped are delivered to a commercial carrier. The recipient is notified to expect delivery; thus, tracking procedures can be initiated quickly in the event samples are not received. Tracking forms and chain-of-custody forms are completed for all transfers of samples maintained by the labs, with copies also maintained by the field team. The information coordinator maintains a centralized tracking system of all shipments.

The field operations phase is completed with collection of all samples or expiration of the sampling window. Following completion of all sampling, a debriefing session will be scheduled (see Table 1-1). These debriefings cover all aspects of the field program and solicit suggestions for improvements.

### *1.3.2 Overview of Laboratory Operations*

Holding times for samples vary with the sample types and analytes. Thus, some analytical analyses (e.g., water chemistry) begin as soon as sampling begins while others are not even initiated until sampling has been completed (e.g., benthic macroinvertebrates). Analytical methods are summarized in the Laboratory Methods Manual that is a companion document to this QAPP. When available, standard methods are used and are referenced. Where experimental methods are used or standard methods are modified, these methods are documented in the laboratory methods manual or in internal documentation, and may be described in SOPs developed by the analytical laboratories.

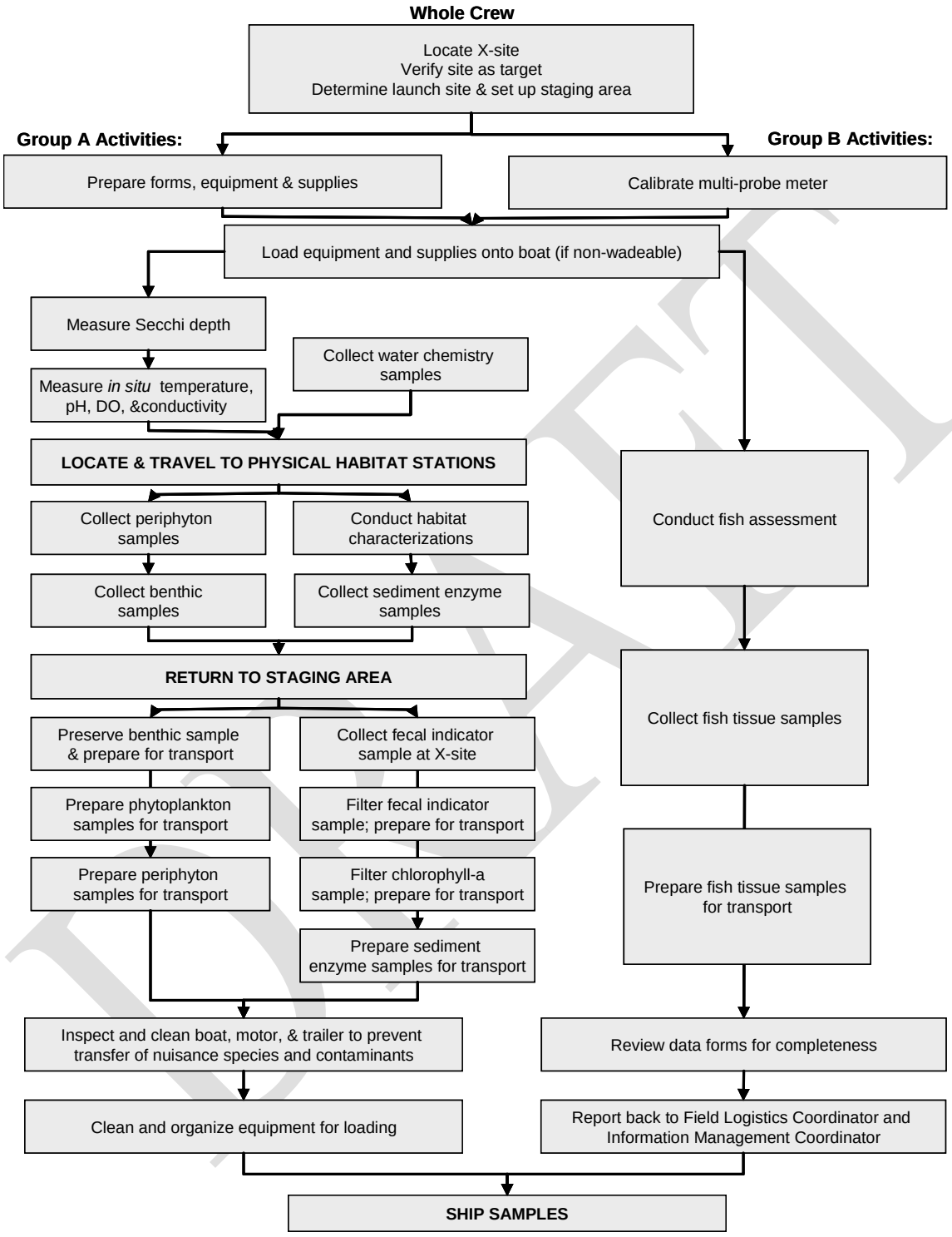


**Figure 4a.** Summary of field activities for boatable stream and river sampling.

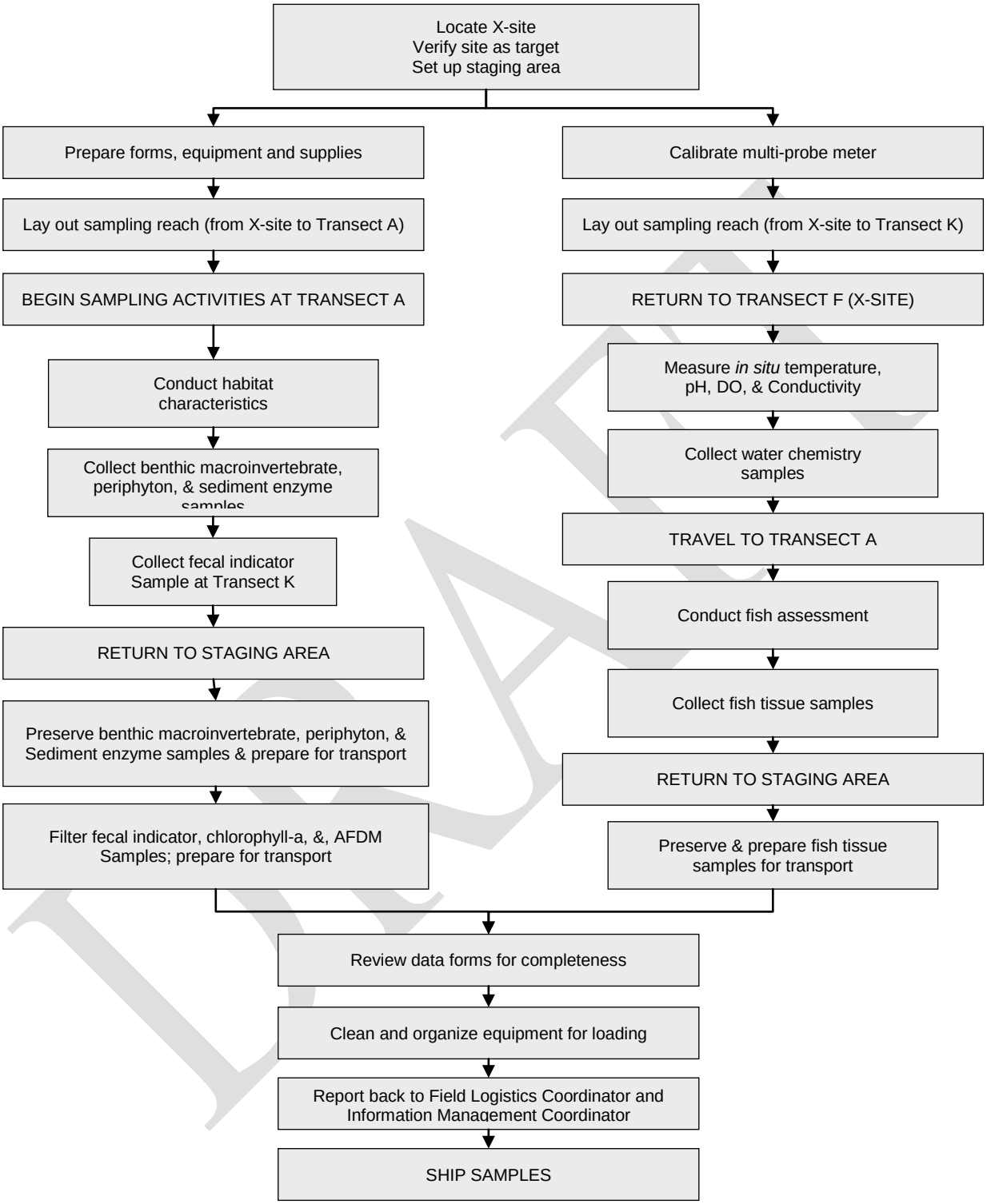


**Figure 4b.** Summary of field activities for wadeable stream sampling.

Water chemistry and chlorophyll-a samples will be analyzed by the contract laboratory, Dynamac, maintained by ORD Western Ecology Division. Benthic macroinvertebrate samples will be processed by a national contractor and a few pre-approved state laboratories. Sediment enzyme and periphyton APA samples will be analyzed by the EPA's National Health and Environmental Effects Research Laboratory in Duluth, MN (NHEERL-Dul). Periphyton ID samples will be analyzed by both the Philadelphia Academy of Natural Sciences and Michigan State University and the state of Wisconsin. Enterococci samples will be analyzed by the EPA's New England Regional Laboratory (NERL). Fish tissue samples will be analyzed by the EPA's National Exposure Research Laboratory in Cincinnati, OH (NERL-Cin). Fish identification vouchers will be verified by the Philadelphia Academy of Natural Sciences and Oregon State University. The physical habitat measurements are made in the field and recorded on the field data sheets and then scanned into a database at the information management center at ORD Western Ecology Division. Laboratories providing analytical support must have the appropriate facilities to properly store and prepare samples, and appropriate instrumentation and staff to provide data of the required quality within the time period dictated by the project. Laboratories must conduct operations using approved laboratory practices (Table 1-2).

All laboratories providing analytical support to the NRSA (water chemistry, chlorophyll a, fish tissue, fish community, benthic macroinvertebrates, sediment enzymes, enterococci, and periphyton) must adhere to the provisions of this integrated QAPP and NRSA Laboratory Manual. Laboratories will provide information documenting their ability to conduct the analyses with the required level of data quality. Such information will include results from interlaboratory comparison studies, analysis of performance evaluation samples, control charts and results of internal QC sample or internal reference sample analyses to document achieved precision, bias, accuracy, and method detection limits. Contracted laboratories will be required to provide copies of their SOPs and audit reports. Water chemistry laboratories may also be required to successfully analyze at least one performance evaluation sample for target analytes before routine samples can be analyzed. Laboratory operations will be evaluated by technical systems audits, performance evaluation studies, and by participation in interlaboratory sample exchange.

**Table 1-2.** Guidelines for analytical support laboratories

| |
|---|
| A program of scheduled maintenance of analytical balances, water purification systems, microscopes, laboratory equipment, and instrumentation. |
| Checking and recording the composition of fresh calibration standards against the previous lot. Acceptable comparisons are ± 2 percent of the theoretical value. |
| Recording all analytical data in bound logbooks in ink, or on standardized recording forms. |
| Monitoring and recording (in a logbook or on a recording form) temperatures and performance of cold storage areas and freezer units. During periods of sample collection operations, monitoring must be done on a daily basis. |
| Verifying the efficiency of fume hoods. |
| If needed, having a source of reagent water meeting American Society of Testing and Materials (ASTM) Type I specifications for conductivity (< 1 :S/cm at 25 /C; ASTM 1984) available in sufficient quantity to support analytical operations. |
| Appropriate microscopes or other magnification for biological sample sorting and organism identification. |
| Labeling all containers used in the laboratory with date prepared, contents, and initials of the individual who prepared the contents. |
| Dating and storing all chemicals safely upon receipt. Chemicals are disposed of properly when the expiration date has expired. |
| Using a laboratory information management system to track the location and status of any sample received for analysis. |
| Reporting results using standard formats and units compatible with the information management system. |

### *1.3.3. Data Analysis and Reporting*

A technical workgroup convened by and under the leadership of the EPA Project Leader is responsible for outlining the final assessment report. Data analysis to support this report will be conducted by the EMAP team at the Western Ecology Division and other experts.. Information management activities in support of this effort are discussed further in Section 4. Data in the database are available to Cooperators for their own use upon completion of the final verification and validation. The final data from the NRSA will be transferred to the OW STORET system.

# 2.0 DATA QUALITY OBJECTIVES

It is a policy of the U.S. EPA and its laboratories that Data Quality Objectives (DQOs) be developed for all environmental data collection activities. Data quality objectives are statements that describe the level of uncertainty that can be associated with environmental data for their intended use. Data quality objectives thus provide the criteria to design a sampling program within cost and resource constraints or technology limitations imposed upon a project or study.

## 2.1 Data Quality Objectives for the NRSA

Target DQOs established for the NRSA relate to the goal of describing the current status in the condition of selected indicators of the condition of rivers and streams in the conterminous U.S. and subregions of interest. The formal statement of the DQO for national estimates is as follows:

Estimate the proportion of river and stream length (± 5%) in the conterminous U.S. that falls below the designated threshold for good conditions for selected measures with 95% confidence.

For the subregions of interest (Omernik Level II Ecoregions) the DQO is:

Estimate the proportion of river and stream length (± 15%) in a specific Level II Ecoregion that falls below the designated threshold for good conditions for selected measures with 95% confidence.

## 2.2 Measurement Quality Objectives

For each indicator, performance objectives (associated primarily with measurement error) are established for several different attributes of data quality (following Smith et al., 1988). Specific objectives for each indicator are presented in the indicator section of this QAPP. The following sections define the data quality attributes and present approaches for evaluating them against acceptance criteria established for the program.

### *2.2.1 Method Detection Limits*

For chemical measurements, requirements for the method detection limit (MDL) are established. The MDL is defined as the lowest level of analyte that can be distinguished from zero with 99% confidence based on a single measurement (1) (Glaser et al., 1981). The MDL for an individual analyte is calculated as:

$$MDL = t_{[\alpha = 0.01,\ \nu = n-1]} \times s$$

where *t* is a Students' *t* value at a significance level (") of 0.01 and n-1 degrees of freedom (<), and *s* is the standard deviation of a set of *n* measurements of a standard solution. The standard contains analyte concentrations between two and three times the MDL objective, and is subjected to the entire analytical method (including any preparation

or processing stages). At least seven non-consecutive replicate measurements are required to calculate a valid estimate of the MDL. Replicate analyses of the standard should be conducted over a period of several days (or several different calibration curves) to obtain a long-term (among-batch) estimate of the MDL.

Laboratories should periodically monitor MDLs on a per batch basis. Suggested procedures for monitoring MDLs are: (1) to analyze a set of serial dilutions of a low level standard, determining the lowest dilution that produces a detectable response; and (2) repeated analysis (at least seven measurements) of a low-level standard within a single batch.

Estimates of MDLs (and how they are determined) are required to be submitted with analytical results. Analytical results associated with MDLs that exceed the detection limit objectives are flagged as being associated with an unacceptable MDL. Analytical data that are below the estimated MDL are reported, but are flagged as being below the MDL.

### *2.2.2 Sampling Precision, Bias, and Accuracy*

Precision and bias are estimates of random and systematic error in a measurement process (Kirchmer, 1983; Hunt and Wilson, 1986). Collectively, precision and bias provide an estimate of the total error or uncertainty associated with an individual measurement or set of measurements. Systematic errors are minimized by using validated methodologies and standardized procedures. Precision is estimated from repeated measurements of samples. Net bias is determined from repeated measurements of solutions of known composition, or from the analysis of samples that have been fortified by the addition of a known quantity of analyte. For analytes with large ranges of expected concentrations, objectives for precision and bias are established in both absolute and relative terms, following the approach outlined in Hunt and Wilson, 1986. At lower concentrations, objectives are specified in absolute terms. At higher concentrations, objectives are stated in relative terms. The point of transition between an absolute and relative objective is calculated as the quotient of the absolute objective divided by the relative objective (expressed as a proportion, e.g., 0.10 rather than as a percentage, e.g., 10%). Final estimates will be calculated by the analysis staff at WED.

Precision in absolute terms is estimated as the sample standard deviation when the number of measurements is greater than two:

$$SD = \sqrt{\frac{i = \sum_{1}^{n} \left(xi - \bar{x}\right)^2}{n-1}}$$

where

$x$ is the value of the replicate

$\bar{X}$ is the mean of repeated sample measurements,

and $n$ is the number of replicates.

Relative precision for such measurements is estimated as the relative standard deviation

(RSD, or coefficient of variation, [CV]):

$$RSD = \frac{s}{\bar{x}} \times 100$$

where

*s* is the sample standard deviation of the set of measurements,

and $\bar{x}$ equals the mean value for the set of measurements.

Precision based on duplicate measurements is estimated based on the range of measured values (which equals the difference for two measurements). The relative percent difference (RPD) is calculated as:

$$RPD = \left( \frac{|A - B|}{A + B} \right) \times 100$$

where

*A* is the first measured value,

*B* is the second measured value.

Precision objectives based on the range of duplicate measurements can be calculated as:

$$Critical\ \ Range = s \times \sqrt{2}$$

where

*s* represents the precision objective in terms of a standard deviation.

Range-based objectives are calculated in relative terms as:

$$Critical\ \ RPD\ = RSD \times \sqrt{2}$$

where

*RSD* represents the precision objectives in terms of a relative standard deviation.

For repeated measurements of samples of known composition, net bias (*B*) is estimated in absolute terms as:

$$B = \bar{x} - T$$

where

$\bar{x}$ equals the mean value for the set of measurements

and *T* equals the theoretical or target value of a performance evaluation sample.

Bias in relative terms (*B[%]*) is calculated as:

$$B(\%) = \frac{\overline{X} - T}{T} \times 100$$

where

$\bar{x}$ equals the mean value for the set of measurements,

and *T* equals the theoretical or target value of a performance evaluation sample.

Accuracy is estimated for some analytes from fortified or spiked samples as the percent recovery. Percent recovery is calculated as:

$$\%\ recovery = \frac{C_{is} - C_i}{C_s} \times 100$$

where

$C_{is}$ is the measured concentration of the spiked sample,

$C_i$ is the concentration of the unspiked sample, and

$C_s$ is the concentration of the spike.

### *2.2.3 Taxonomic Precision and Accuracy*

For the NRSA, taxonomic precision will be quantified by comparing whole-sample identifications completed by independent taxonomists or laboratories. Accuracy of taxonomy will be qualitatively evaluated through specification of target hierarchical levels (e.g., family, genus, or species); and the specification of appropriate technical taxonomic literature or other references (e.g., identification keys, voucher specimens). To calculate taxonomic precision, 10% of the biological samples from each participating laboratory will be randomly-selected by EPA HQ, and sent to an independent taxonomist for re-identification. Comparison of the results of whole sample re-identifications will provide a Percent Taxonomic Disagreement (PTD) calculated as:

$$PTD = \left[1 - \left(\frac{comp_{pos}}{N}\right)\right] \times 100$$

where $comp_{pos}$ is the number of agreements, and $N$ is the total number of individuals in the larger of the two counts. The lower the PTD, the more similar are taxonomic results and the overall taxonomic precision is better. A measurement quality objective (MQO) of 15% is recommended for taxonomic difference or disagreement (overall mean ≤ 15% is acceptable based on similar projects)for benthic macroinvertebrates and fish. Individual samples exceeding 15% are examined for taxonomic areas of substantial disagreement, and the reasons for disagreement investigated. Periphyton and algal samples have a higher PTD due to the variance amongst species.

Sample enumeration is another component of taxonomic precision. Sample enumeration agreement will be checked with the same 10% of samples used to check taxonomic precision. Final specimen counts for samples are dependent on the taxonomist, not the rough counts obtained during the sorting activity. Comparison of counts is quantified by calculation of percent difference in enumeration (PDE), calculated as:

$$PDE = \left(\frac{|Lab1 - Lab2|}{Lab1 + Lab2}\right) \times 100$$

An MQO of 5% is recommended (overall mean of ≤ 5% is acceptable) for several biological

samples, while others will have higher PDE's. This is based on the laboratory approaches used and the nature of the indicator. Specific PDE's are in each indicator section.

Corrective actions for samples exceeding these MQOs can include defining the taxa for which re-identification may be necessary (potentially even by third party), for which samples (even outside of the 10% lot of QC samples) it is necessary, and where there may be issues of nomenclatural or enumeration problems. Taxa lists will be changed when disagreements are resolved by a third party.

Taxonomic accuracy is evaluated by having individual specimens representative of selected taxa identified by recognized experts, usually contract or university affiliated persons who have peer-reviewed publications for the taxonomic group they are reviewing. Samples will be identified using the most appropriate technical literature that is accepted by the taxonomic discipline and reflects the accepted nomenclature. The Integrated Taxonomic Information System (ITIS, http://www.itis.usda.gov/) will be used to verify nomenclatural validity and reporting. A reference collection will be compiled by each lab as the samples are identified. Specialists in several taxonomic groups will verify selected individuals of different taxa, as determined by the NRSA workgroup.

### *2.2.4 Completeness*

Completeness requirements are established and evaluated from two perspectives. First, valid data for individual indicators must be acquired from a minimum number of sampling locations in order to make subpopulation estimates with a specified level of confidence or sampling precision. The objective of this study is to complete sampling at 95% or more of the 1800 initial sampling sites and the 200 reference sites. Percent completeness is calculated as:

$$\%C = V / T \times 100$$

where V = number of measurements/samples judged valid, and T = total number of planned measurements/samples. Within each indicator, completeness objectives are also established for individual samples or individual measurement variables or analytes. These objectives are estimated as the percentage of valid data obtained versus the amount of data expected based on the number of samples collected or number of measurements conducted. Where necessary, supplementary objectives for completeness are presented in the indicator-specific sections of this QAPP.

### *2.2.5 Comparability*

Comparability is defined as the confidence with which one data set can be compared to another (Stanley and Verner, 1985; Smith et al., 1988). For all indicators, comparability is addressed by the use of standardized sampling procedures, sampling equipment and analytical methodologies by all sampling crews and laboratories. These are also the same used to collect data in EMAP West and WSA studies. Comparability of data within and among indicators is also facilitated by the implementation of standardized quality

assurance and quality control techniques and standardized performance and acceptance criteria. For all measurements, reporting units and format are specified, incorporated into standardized data recording forms, and documented in the information management system. Comparability is also addressed by providing results of QA sample data, such as estimates of precision and bias, conducting methods comparison studies when requested by the grantees and conducting interlaboratory performance evaluation studies among state, university, and NRSA contract laboratories. If some incompatibility between sampling crews comes to light, the data will be rejected.

### *2.2.6 Representativeness*

Representativeness is defined as "the degree to which the data accurately and precisely represent a characteristic of a population parameter, variation of a property, a process characteristic, or an operational condition" (Stanley and Verner, 1985, Smith et al., 1988). At one level, representativeness is affected by problems in any or all of the other attributes of data quality.

At another level, representativeness is affected by the selection of the target surface water bodies, the location of sampling sites within that body, the time period when samples are collected, and the time period when samples are analyzed. The probability-based sampling design should provide estimates of condition of surface water resource populations that are representative of the region. The individual sampling programs defined for each indicator attempt to address representativeness within the constraints of the sampling design and index sampling period. Holding time requirements for analyses ensure analytical results are representative of conditions at the time of sampling. Use of QC samples which are similar in composition to samples being measured provides estimates of precision and bias that are applicable to sample measurements.

## 3.0 SURVEY DESIGN

Many of the questions which USEPA's Office of Water, States and Tribes are attempting to address fundamentally require information about large numbers of systems rather than individual systems. ORD has studied the role of monitoring surveys, their evolution and the nature of existing federal monitoring programs, and can provide information and assistance to the States and Tribes in this area.

The survey design for the NRSA is the same as used for EMAP-West plus the Great Rivers and the tidal systems. The design is a sample survey design (a.k.a. probability design) that ensures a representative set of sample sites from which inferences can be made about the target population. For the NRSA, the target population is all National rivers and streams in the conterminous US, excluding sites below the head of salt or reservoirs.

There is a large body of statistical literature dealing with sample survey designs which addresses the problem of making statements about many by sampling the few (e.g., Cochran 1977, Kish 1965, Kish 1987, Sarndal et al. 1992). Sample surveys have been used in a variety of fields (e.g., election polls, monthly labor estimates, forest inventory analysis, national wetlands inventory) to determine the status of populations (large groups of sites) of interest, especially if the population is too numerous to census or if it is unnecessary to census the population to reach the desired level of precision for describing the population's status. A key point in favor of probability based designs is that they allow lower cost sampling programs because a smaller number of sites are able to support conclusions with known accuracy and precision about status and trends of a region.

Probability sampling surveys have been consistently used in some natural resource fields. The National Agricultural Statistics Survey (NASS) conducted by the U.S. Department of Agriculture and the Forest Inventory Analysis (FIAT) conducted by the U.S. Forest Service (Bickford et al. 1963, Hazard and Law 1989) have both used probability based sampling concepts to monitor and estimate the condition and productivity of agricultural and forest resources from a commodity perspective. National Resources Inventory (NRI) was instituted initially because of concerns about the impact of soil erosion on crop production. More recently, the National Wetland Inventory (NWI) developed by the U.S. Fish and Wildlife Service (Wilen 1990) to estimate the extent of wetland acreage in the United States has used a probability based sampling design. However, no thorough review of all national programs has occurred until recently.

The survey designs used in EMAP to date have been documented in published reports for each resource group and in the peer reviewed literature. Below a brief description of the design concepts and the specific application for riverine systems is provided. Much of this is extracted from various publications and from Stevens (1994) which provides an excellent overview of the design concepts, issues and applications for the entire program. The EMAP sampling design strategy is based on the fundamental requirement for a probability sample of an explicitly defined regional resource population, where the sample is constrained to reflect the spatial dispersion of the population.

A key property of a probability sample is that every element in the population has some chance of being included in the sample. If this were not the case, then some parts of the population might as well not exist, since no matter what, their condition could have no influence on estimates of population characteristics. This property has a side benefit, in that it forces an explicit and complete definition of the population being described. This may seem trivial; however, in practice, it is almost never easy to tightly delimit a real, physical population. For example, "river" is a concept that has meaning for most people, and the notion of "all rivers in the continental United States" would seem to define a population. Nevertheless, an operational definition of membership is missing. The operational definition must be complete enough to establish any flowing water, from a headwater stream up to the Mississippi River, as either in or out of the population. Thus, the definition must address such aspects as size limits (at least lower limits on flow), natural rivers versus constructed channels, temporal fluctuation (If a "river" dries up during a drought, is it still a river? Was it a river before the drought?), and amount of flowing water and riparian zone. Without such an operational definition, any statement about "all rivers in the United States" has an unquantifiable vagueness.

The river and stream resource does not fall neatly into either the discrete or extensive category. The National Stream Survey (Messer et al., 1986; Overton, 1985) split streams into reaches defined as the length of stream between confluences, or from the headwaters down to the first confluence. Thus, streams were treated as a finite discrete population. A grid was used to sample stream reaches by randomly placing a grid over a topographic map of the area of interest, and then proceeding downhill along the fall line until a stream reach was intersected. The approach that was taken avoids the necessity of delimiting the resource areal units. The approach of EMAP-West is somewhat different. The program focuses on the population of stream miles rather than stream reaches. We wish to characterize the population in terms of the condition of length of rivers and streams rather than numbers of river or stream reaches. Therefore, we want a sampling method that samples a river or stream in proportion to its length; this is accomplished by viewing rivers and streams as an extensive resource with length. The method described here is currently being used in a pilot study, which, among other goals, will examine the suitability of the method for a larger study. Stream and river traces are identified on 1:100,000-scale Digital Line Graphs, and a Geographical Information System is used to intersect these with the sampling templates. Each river and stream segment within a template is identified and its length determined. The endpoints of a segment are defined as confluences, headwaters ends, or intersections with a template edge. Sets of connected segments of the same order are always kept together in the sample selection process. The appropriate Strahler stream order is also determined for each segment.

Some differential weighting by size is necessary because of the predominance of lower-order streams. The sample selection proceeds with inclusion probability for a segment proportional to its length times the weight for its order. The total inclusion probability for each template is calculated as the weighted sum of stream lengths in the template, the templates are partitioned into groups using the partitioning algorithm described for lakes, and the samples are selected in an analogous manner: The partitions are randomized, the templates are randomized within the partitions, and the sets of connected segments are randomized within the templates. The same systematic selection protocol is used;

however, in this case, the selection not only identifies the stream segment to be sampled, but also identifies the point on that segment where the sample is to be located. This is accomplished by recording the relative distance from the beginning of the segment to the selected point on the segment.

The types of questions which have been posed from various State and Tribal agencies suggest that they would like to make statements about all streams and rivers. Clearly, sampling every mile of river and stream in the country is not economically feasible nor is it necessary. Probability designs have been used in a wide range of disciplines to address this need (Converse 1987).

The primary objectives of this study are to estimate the condition of mapped perennial National rivers and streams, and the extent (total length) of mapped channels, in conterminous states of the U.S. The objectives specify an interest in the target population of wadeable and non-wadeable perennial streams and rivers.

One estimate of extent is provided by National Hydrography Database Plus (NHD- Plus) which is based on digitized blue lines from 1:100,000 scale maps. Based on prior information, it is known that NHD-Plus incorrectly codes some stream segments. Incorrect code information occurs for (1) designating Strahler stream order; (2) delineating perennial and intermittent, (3) defining natural versus constructed channels, including newly modified channels, and (4) distinguishing irrigation return flow from irrigation delivery channels. In some cases, NHD-Plus includes stream channels that are not actually present, due to (1) no definable channel present, (2) location is wetland/marsh with no defined channel, or (3) channel may be an impoundment. NHD-Plus may also exclude some stream channels due to (1) mapping inconsistencies in construction of 1:100,000 maps, (2) digitization of map blue lines, or (3) inadequacy of photo information used to develop maps, e.g. heavily forested areas with low order streams. This study assumes that NHD-Plus includes all stream channels specified by the definition of the target population. That is, if stream channels exist that are not included in NHD-Plus, they will not be addressed by this study.

A secondary outcome of estimating the extent of the stream channel resource will be estimates on the amount of miscoding present in NHD-Plus. Those stream segments actually selected in the survey sample that are found to be miscoded will be submitted to NHD-Plus staff for correction.

### 3.1 Probability-Based Sampling Design and Site Selection

Target Population: Within the conterminous U.S, all stream and river channels (natural and constructed) mapped at 1:100,000 scale

Sample Frame: NHD-Plus stream and river channel segments coded as R, S, T, N, W, (412, 413, 999) and U (414, 415).

This frame is subdivided into two major parts: (1) all NHD-Plus stream, river and canal segments coded as perennial, and (2) all NHD-Plus stream, river and canal segments coded as non-perennial, i.e., all other stream, river and canal segments. The purpose of

subdividing the frame is to allow a sampling focus on systems that have an exceedingly high probability of being flowing waters during the index sampling period.

Sites were selected for the NRSA project using a hierarchical randomization design process described by Stevens and Olsen (1999, 2003, 2004). The national hydrography database (NHD) served as the frame representing streams and rivers in the US. Data from approximately 1800 river and stream sites in the United States will be used in the assessment and sampled over a two year index period. This total sample size will allow national reporting as well as regional reporting at the scale of 9 aggregated Omernik Level II ecoregions, the ten EPA Regions and 10-15 major drainage basins. Several States have added additional sites to be able to report on the condition of streams and/or rivers within their boundaries.

Key features of the approach are (1) utilizing survey theory for continuous populations within a bounded area, (2) explicit control of the spatial dispersion of the sample through hierarchical randomization, (3) unequal probability of selection by Strahler order, and (4) nested subsampling to incorporate intensified sampling in special study regions.

Revisit Sites: Of the sites visited in the field and found to be target sites, a total of 10% will be revisited. The 10% will be the first 10% of the sites visited. The primary purpose of this revisit set of sites is to allow variance estimates that would provide information on the extent to which the population estimates might vary. In addition 450 WSA streams will be revisited during the 2008 and 2009 sampling season to evaluate change from the WSA.

Site Evaluation Sites: The number of sites that must be evaluated to achieve the expected number of field sites that can be sampled can only be estimated based on assumptions concerning expected error rates in RF3, percent of landowner refusals, and percent of physically inaccessible sites. Based on the estimates gained in previous studies, a list of alternate sites was selected at the same time as the base sites. These alternate sites will be using in order until the desired sample designated for the state has been acheived.

# 4.0 INFORMATION MANAGEMENT

Like QA, information management (IM) is integral to all aspects of the NRSA from initial selection of sampling sites through dissemination and reporting of final, validated data. QA and QC measures implemented for the IM system are aimed at preventing corruption of data at the time of their initial incorporation into the system and maintaining the integrity of data and information after incorporation into the system. The general organization of, and QA/QC measures associated with, the IM system are described in this section.

Long-term data from the NRSA will be maintained in STORET/WQX and the EMAP data system at ORD (formerly Surface Water Information Management System) . Project data management activities will be handled at EPA's Western Ecology Division and will be compliant with all relevant EPA and Federal data standards. Data will be shipped from sample processing laboratories to WED no later than May 2011.

## 4.1 Data Policy

The NRSA requires a continuing commitment to the establishment, maintenance, description, accessibility, and long-term availability of high-quality data and information. All data used in the NRSA will be maintained, following final verification and validation of dataset, in EPA's STORET/WQX and EPA's EMAP data system.

Full and open sharing of the full suite of data and published information produced by the study is a fundamental objective. Data and information will be available without restriction for no more than the cost of reproduction and distribution. Where possible, the access to the data will be via the World Wide Web through STORET and EMAP to keep the cost of delivery to a minimum and to allow distribution to be as wide as possible. All data collected by this study will be publicly available following verification and validation of the dataset.

Organizations and individuals participating in the project will ship all samples in a timeline consistent with the field operations manual. Field data sheets will be sent directly to WED for data entry. All laboratories processing samples will send final electronic dataset to WED by May 2011. Data and metadata will be available for assessment preparation by July 2010. Final dataset with metadata will be available via STORET and EMAP at the time of delivery of the final report, December 2011.

All data sets and published information used in the study will be identified with a citation; for data sets an indication of how the data may be accessed will be provided. Data from this study will be maintained indefinitely. All EPA data policies will be followed including EPA data standards, GIS, etc., as discussed in section 4.3.

## 4.2 Overview of System Structure

At each point where data and information are generated, compiled, or stored, the information must be managed. Thus, the IM system includes all of the data-generating activities, all of the means of recording and storing information, and all of the processes which use

data. The IM system includes both hardcopy and electronic means of generating, storing, and archiving data. All participants in the NRSA have certain responsibilities and obligations which make them a part of the IM system. In its entirety, the IM system includes site selection and logistics information, sample labels and field data forms, tracking records, map and analytical data, data validation and analysis processes, reports, and archives. IM staff supporting the NRSA at WED provide support and guidance to all program operations in addition to maintaining a central data base management system for the NRSA data.

The central repository for data and associated information collected for use by the NRSA is a DEC Alpha server system located at WED-Corvallis. The general organization of the information management system is presented in Figure 5. Data are stored and managed on this system using the Statistical Analysis System (SAS) software package. This centrally managed IM system is the primary data management center for the NRSA research conducted at WED and elsewhere. The IM staff receives, enters, and maintains data and information generated by the site selection process (see Section 3), field sample and data collection, map-based measurements, laboratory analyses, and verification and validation activities completed by the states, cooperators and contractors. In addition to this inflow, the IM system provides outflow in provision of data files to NRSA staff and other users. The IM staff at WED is responsible for maintaining the security integrity of both the data and the system.

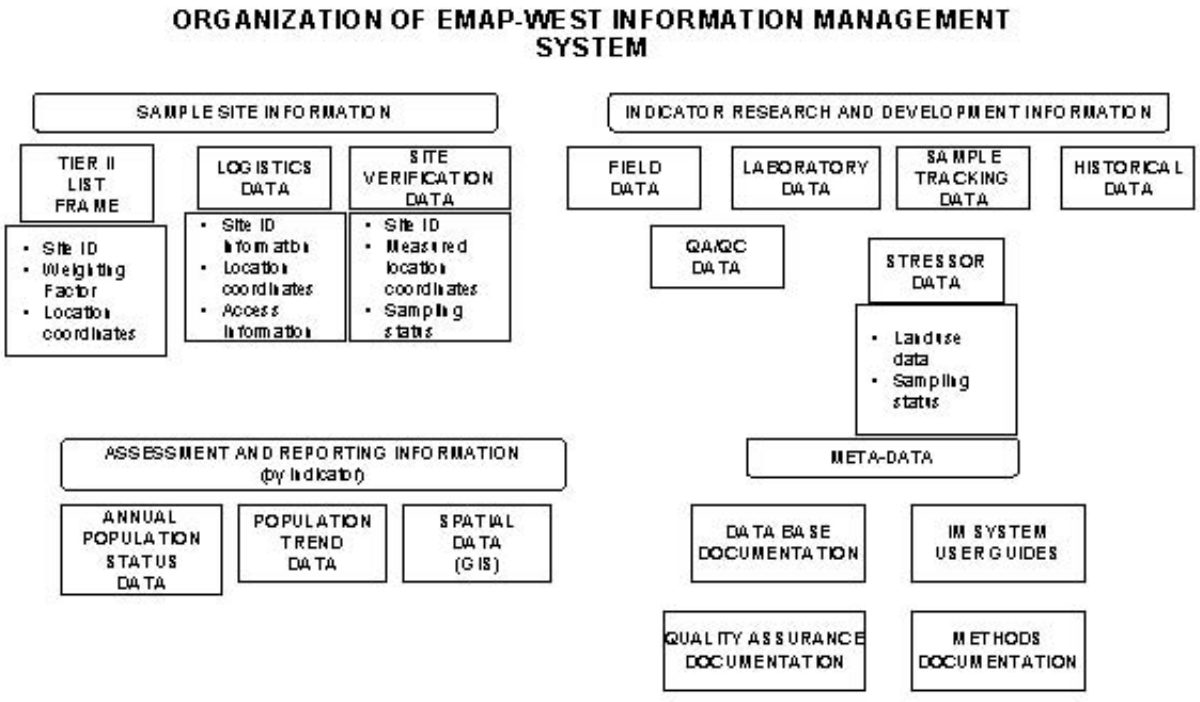


**Figure 5.** Organization of information management system modeled after EMAP-WEST for the NRSA.

The following sections describe the major inputs to the central data base and the associated QA/QC processes used to record, enter, and validate measurement and analytical data collected for EMAP surface waters research projects. Activities to maintain the integrity and assure the quality of the contents of the IM system are also described.

### *4.2.1 Design and Logistics Data Bases*

The site selection process described in Section 3 produces a list of candidate sampling locations, inclusion probabilities, and associated site classification data (e.g., target status, ecoregion, stream order, etc.). This "design" data base is provided to the IM staff, implementation coordinators, and field coordinators. Field coordinators determine ownership and contacts for acquiring permission to access each site, and conduct reconnaissance activities. Ownership and reconnaissance information for each site are compiled into a "logistics" data base. Generally, standardized forms are used during reconnaissance activities. Information from these forms may be entered into a SAS compatible data management system. Whether in electronic or hardcopy format, a copy of the logistics data base is provided to the IM for archiving storage.

### *4.2.2 Sample Collection and Field Data Recording*

Prior to initiation of field activities, the IM staff develops standardized field data forms and sample labels. Preprinted adhesive labels having a standard recording format are completed and affixed to each sample container. Precautions are taken to ensure that label information remains legible and the label remains attached to the sample. Examples of sample labels are presented in the field operations manual.

Field sample collection and data forms are designed in conjunction with IM staff to ensure the format facilitates field recording and subsequent data entry tasks. All forms which may be used onsite are printed on water-resistant paper. Copies of the field data forms and instructions for completing each form are documented in the field operations manuals. Recorded data are reviewed upon completion of data collection and recording activities by a person other than the one who completed the form. Field crews check completed data forms and sample labels before leaving a sampling site to ensure information and data were recorded legibly and completely. Errors are corrected if possible, and data considered as suspect are qualified using a flag variable. The field crew enters explanations for all flagged data in a comments section. Completed field data forms are transmitted to the IM staff at WED for entry into the central data base management system.

All samples are tracked from the point of collection. Hardcopy tracking and custody forms are completed by the field crews. Copies of the shipping and custody record accompany all sample transfers; other copies are transmitted to the IMC and applicable indicator lead. Samples are tracked to ensure that they are delivered to the appropriate laboratory, that lost shipments can be quickly identified and traced, and that any problems with samples observed when received at the laboratory are reported promptly so that corrective action

can be taken if necessary. Detailed procedures on shipping and sample tracking can be found in Appendix C of the Field Operations Manual

Procedures for completion of sample labels and field data forms, and use of PCs are covered extensively in training sessions. General QC checks and procedures associated with sample collection and transfer, field measurements, and field data form completion for most indicators are listed in Table 3-1. Additional QA/QC checks or procedures specific to individual indicators are described in the indicator sections in Section 5 of this QAPP.

### *4.2.3 Laboratory Analyses and Data Recording*

Upon receipt of a sample shipment, analytical laboratory receiving personnel check the condition and identification of each sample against the sample tracking record. Each sample is identified by information written on the sample label and by a barcode label. Any discrepancies, damaged samples, or missing samples are reported to the IM staff and indicator lead by telephone. The laboratory receiving personnel log in the samples and post the log-in information for the IM staff at WED, who track all sample shipping, custody, and disposition.

**Table 4-1.** Sample and field data quality control activities

| Quality Control Activity | Description and/or Requirements |
|---|---|
| Contamination Prevention | All containers for individual site sealed in plastic bags until use; specific contamination avoidance measures covered in training |
| Sample Identification | Pre-printed labels with unique ID number for each sample |
| Data Recording | Data recorded on pre-printed forms of water-resistant paper; field crew reviews data forms for accuracy, completeness, and legibility |
| Data Qualifiers | Defined qualifier codes used on data form; additional qualifiers explained in comments section on data form |
| Sample Custody | Unique sample ID and tracking form information entered in an electronic laboratory information management system (LIMS); sample shipment and receipt confirmed |
| Sample Tracking | Sample condition inspected upon receipt and noted on tracking form with copies sent to Indicator Lead, Communications Center, and/or IM |
| Data Entry | Data entered using customized entry screens that resemble the data forms; entries reviewed manually or by automated comparison of double entry |
| Data Submission | Standard format defined for each measurement including units, significant figures, and decimal places, accepted code values, and required field width |
| Data Archival | All data archived in an organized manner for a period of seven years or until written authorization for disposition has been received from the Surface Waters Technical Director. |

Most of the laboratory analyses for the NRSA indicators, particularly chemical and physical analyses, follow or are based on standard methods. Standard methods generally include requirements for QC checks and procedures. General laboratory QA/QC procedures applicable to most NRSA indicators are described in Table 4-2. Additional QA/QC samples and procedures specific to individual indicator analyses are described in the indicator sections in Part II of this QAPP. Biological sample analyses are generally based on current acceptable practices within the particular biological discipline. Some QC checks and procedures applicable to most NRSA biological samples are described in Table 4-3. Additional QA/QC procedures specific to individual biological indicators are described in the indicator sections in Part 5 of this QAPP.

A laboratory's IM system may consist of only hardcopy records such as bench sheets and logbooks, an electronic laboratory information management system (LIMS), or some combination of hardcopy and electronic records. Laboratory data records are reviewed at the end of each analysis day by the designated laboratory onsite QA coordinator or by supervisory personnel. Errors are corrected if possible, and data considered as suspect by laboratory analysts are qualified with a flag variable. All flagged data are explained in a comments section. Private contract laboratories generally have a laboratory quality assurance plan and established procedures for recording, reviewing, and validating analysis data. Once analytical data have passed all of the laboratory's internal review procedures, a submission package is prepared and transferred to the IM staff. The contents of the submission package are largely dictated by the type of analysis (physical, chemical, or biological), but generally includes at least the elements listed in Tables 4-2 or 4-3. All samples and raw data files (including logbooks, bench sheets, and instrument tracings) are to be retained for a period of seven years or until authorized for disposal, in writing, by the NRSA Project Leader.

**Table 4-2.** Laboratory data quality control activities

| Quality Control Activity | Description and/or Requirements |
|---|---|
| Instrument Maintenance | Follow manufacturer's recommendations and specific guidelines in methods; maintain logbook of maintenance/repair activities |
| Calibration | Calibrate according to manufacturer's recommendations and guidelines given in Section 6; recalibrate or replace before analyzing any samples |
| QC Data | Maintain control charts, determine MDLs and achieved data attributes; include QC data summary in submission package |
| Data Recording | Use software compatible with EMAP-SWIM system; check all data entered against the original bench sheet to identify and correct entry errors. Review other QA data (e.g. condition upon receipt, etc.) for possible problems with sample or specimens. |
| Data Qualifiers | Use defined qualifier codes; explain all additional qualifiers |
| Data Entry | Automated comparison of double entry or 100% manual check against original data form |

| | |
|---|---|
| Submission Package | Includes: Letter by the laboratory manager; data, data qualifiers and explanations; electronic format compatible with EMAP-SWIM system, documentation of file and data base structures, variable descriptions and formats; summary report of any problems and corrective actions implemented |

**Table 4-3.** Biological sample quality control activities

| Quality Control Activity | Description and/or Requirements |
|---|---|
| Sorting/Enumeration | Re-sort 10% of samples and check counts of organisms |
| Taxonomic Nomenclature | Use accepted common and scientific nomenclature and unique entry codes |
| Taxonomic Identifications | Use standard taxonomic references and keys; maintain bibliography of all references used |
| Independent Identifications | Uncertain identifications to be confirmed by expert in particular taxa |
| Duplicate Identifications | At least 5% of all samples completed per taxonomist reidentified by different analyst; less than 15% assigned different ID |
| Taxonomic Reasonableness Checks | Species or genera known to occur in given conditions or geographic area |

### *4.2.4 Data Review, Verification, Validation Activities*

Raw data files are created from entry of field and analytical data, including data for QA/QC samples and any data qualifiers noted on the field forms or analytical data package. After initial entry, data are reviewed for entry errors by either a manual comparison of a printout of the entered data against the original data form or by automated comparison of data entered twice into separate files. Entry errors are corrected and reentered. For biological samples, species identifications are corrected for entry errors associated with incorrect or misspelled codes. Errors associated with misidentification of specimens are corrected after voucher specimens have been confirmed and the results are available. Files corrected for entry errors are considered to be raw data files. Copies of all raw data files are maintained in the centralized IM system.

Some of the typical checks made in the processes of verification and validation are described in Table 4-4. Automated review procedures may be used. The primary purpose of the initial checks is to confirm that a data value present in an electronic data file is accurate with respect to the value that was initially recorded on a data form or obtained from an analytical instrument. In general, these activities focus on individual variables in the raw data file and may include range checks for numeric variables, frequency tabulations of coded or alphanumeric variables to identify erroneous codes or misspelled entries, and summations of variables reported in terms of percent or percentiles. In addition, associated QA information (e.g., sample holding time) and QC sample data are reviewed to determine if they meet acceptance criteria. Suspect values are assigned a data qualifier until they can be corrected or confirmed as unacceptable and replaced with a

new acceptable value from sample reanalysis.

**Table 4-4.** Data review, verification, and validation quality control activities

| Quality Control Activity | Description and/or Requirements |
|---|---|
| Review any qualifiers associated with variable | Determine if value is suspect or invalid; assign validation qualifiers as appropriate |
| Summarize and review replicate sample data | Identify replicate samples with large variance; determine if analytical error or visit-specific phenomenon is responsible |
| Determine if data quality objectives have been achieved | Determine potential impact on achieving research and/or program objectives |
| Exploratory data analyses (univariate, bivariate, multivariate) utilizing all data | Identify outlier values and determine if analytical error or site-specific phenomenon is responsible |
| Confirm assumptions regarding specific types of statistical techniques being utilized in development of metrics and indicators | Determine potential impact on achieving research and/or program objectives |

A second review is conducted after all analyses have been completed and the raw data file is created. The internal consistency among different analyses or measurements conducted on a sample is evaluated. Examples of internal consistency checks include calculation of chemical ion balances or the summation of the relative abundances of taxa. Samples identified as suspect based on internal consistency checks are qualified with a flag variable and targeted for more intensive review. Data remain qualified until they can be corrected, are confirmed as acceptable in spite of the apparent inconsistency, or until new acceptable values are obtained from sample reanalysis. Upon completion of these activities, copies of the resultant data files are transmitted for archival storage.

In the final stage of data verification and validation, exploratory data analysis techniques may be used to identify extreme data points or statistical outliers in the data set. Examples of univariate analysis techniques include the generation and examination of box-and-whisker plots and subsequent statistical tests of any outlying data points. Bivariate techniques include calculation of Spearman correlation coefficients for all pairs of variables in the data set with subsequent examination of bivariate plots of variables having high correlation coefficients. Recently, multivariate techniques have been used in detecting extreme or outlying values in environmental data sets (Meglen, 1985; Garner et al., 1991; Stapanian et al., 1993). A software package, SCOUT, developed by EPA and based on the approach of Garner et al. (1991) may be used for validation of multivariate data sets.

Suspect data are reviewed to determine the source of error, if possible. If the error is correctable, the data set is edited to incorporate the correct data. If the source of the error cannot be determined, data are qualified as questionable or invalid. Data qualified

as questionable may be acceptable for certain types of data analyses and interpretation activities. The decision to use questionable data must be made by the individual data users. Data qualified as invalid are considered to be unacceptable for use in any analysis or interpretation activities and will generally be removed from the data file and replaced with a missing value code and explanatory comment or flag code. After completion of verification and validation activities, a final data file is created, with copies transmitted for archival and for uploading to the centralized IM system.

Once verified and validated, data files are made available for use in various types of interpretation activities, each of which may require additional restructuring of the data files. These restructuring activities are collectively referred to as "data enhancement." In order to develop indicator metrics from one or more variables, data files may be restructured so as to provide a single record per stream or river site. To calculate site population estimates based on individual measurements or indicators, missing values and suspect data points may need to be replaced with alternate data (such as a value from a replicate measurement) or values calculated from predictive relationships based on other variables.

### 4.3 Data Transfer

Field crews may transmit data electronically via email or CD; original hardcopies of completed data and sample tracking forms must be transmitted to the IM staff at WED via express courier service. Copies of raw, verified, and validated data files are transferred from states, cooperators, and contractors to the IM staff for inclusion in the central IM system. All transfers of data are conducted using a means of transfer, file structure, and file format that has been approved by the IM staff. Data files that do not meet the required specifications will not be incorporated into the centralized data access and management system.

### 4.4 Core Information Management Standards

Participants will adhere to the “Core Information Management Standards for the EMAP Western Study.” National and international standards will be used to the greatest extent possible. This section details a list of standards pertaining to information management that all participants in the NRSA agree to follow. The goal of these core standards is to maximize the ability to exchange data with other studies conducted under the monitoring framework of the Committee on the Environment and Natural Resources (CENR 1997). The main standards are those of the Federal Geographic Data Committee (FGDC 1999), the National Spatial Data Infrastructure (NSDI 1999), and the National Biological Information Infrastructure (NBII 1999).

#### *4.4.1 Metadata*

Federal Geographic Data Committee Content standard for digital geospatial metadata, version 2.0. FGDC-STD-001-1998 (FGDC 1998), including the Biological Data Profile and the Biological Names and Taxonomy Data Standards developed by the National Biological Information Infrastructure (NBII 1999).

For tabular data, metadata that meet the FGDC content standard are contained by a combination of the EMAP Data Directory and the EMAP Data Catalog. For ARC/INFO coverages, the metadata are in the .DOC file embedded in the coverage. This file stays with the coverage. When the coverage is moved to the EMAP public web sites, it will be duplicated to an ASCII text file.

### *4.4.2 Data Directory*

The EMAP Data Directory is maintained as an Oracle database. The guidelines are given in Frithsen and Strebel (1995), Frithsen (1996a, b) and USEPA (1996b).

EMAP Directory entries are periodically uploaded to the Environmental Information Management system (EIMS 1999).. The EIMS will become EPA's node for the National Spatial Data Infrastructure and will make directory information available to other federal agencies through the Z39.50 protocol in accordance with the US Global Change Research Program (USGCRP 1998)

### *4.4.3 Data Catalog*

Data catalog standards are given in Frithsen and Strebel (1995), Frithsen (1996a), and USEPA (1996c).

### *4.4.4 Data Formats*

Attribute data ASCII files: comma-separated values, or space-delimited, or fixed column SAS export files Oracle; GIS data ARC/INFO export files; compressed .tar file of ARC/INFO workspace Spatial Data Transfer Standard (SDTS) (FGDC 1999) format available on request

### *4.4.5 Parameter Formats*

Sampling Site (EPA Locational Data Policy (USEPA 1991)
Latitude and Longitude in decimal degrees (+/- 7.4), Negative longitude values (west of the prime meridian), NAD83
Date: YYYYMMDD (year, month, day)
Hour: HHMMSS (hour, minute, second), Greenwich mean time, Local time

Data loaded to STORET will take on the STORET formats upon loading.

### *4.4.6 Standard Coding Systems*

Chemical Compounds: Chemical Abstracts Service (CAS 1999)
Species Names: Integrated Taxonomic Information system (ITIS 1999)
Land cover/land use codes: Multi-Resolution Land Characteristics (MRLC 1999)

## 4.5 Hardware and Software Control

All automated data processing (ADP) equipment and software purchased for or used in the

NRSA surface waters research is subject to the requirements of the federal government, the particular Agency, and the individual facility making the purchase or maintaining the equipment and software. All hardware purchased by EPA is identified with an EPA barcode tag label; an inventory is maintained by the responsible ADP personnel at the facility. Inventories are also maintained of all software licenses; periodic checks are made of all software assigned to a particular PC.

The development and organization of the IM system is compliant with guidelines and standards established by the EMAP Information Management Technical Coordination Group, the EPA Office of Environmental Information (OEI), and the EPA office of Administrative Resources Management (OARM). Areas addressed by these policies and guidelines include, but are not limited to, the following:

- Taxonomic Nomenclature and Coding
- Locational data
- Sampling unit identification and reference
- Hardware and software
- Data catalog documentation

The NRSA is committed to compliance with all applicable regulations and guidance concerning hardware and software procurement, maintenance, configuration control, and QA/QC. As new guidance and requirements are issued, the NRSA information management staff will assess the impact upon the IM system and develop plans for ensuring timely compliance.

### 4.6 Data Security

All data files in the IM system are protected from corruption by computer viruses, unauthorized access, and hardware and software failures. Guidance and policy documents of EPA and management policies established by the IM Technical Coordination Group for data access and data confidentiality are followed. Raw and verified data files are accessible only to the NRSA collaborators. Validated data files are accessible only to users specifically authorized by the EPA Project Leader. Data files in the central repository used for access and dissemination are marked as read-only to prevent corruption by inadvertent editing, additions, or deletions.

Data generated, processed, and incorporated into the IM system are routinely stored as well as archived on redundant systems. This ensures that if one system is destroyed or incapacitated, IM staff will be able to reconstruct the data bases. Procedures developed to archive the data, monitor the process, and recover the data are described in IM documentation.

Several backup copies of all data files and of the programs used for processing the data are maintained. Backups of the entire system are maintained off-site. System backup procedures are utilized. The central data base is backed up and archived according to procedures already established for WED. All laboratories generating data and developing data files must have established procedures for backing up and archiving

computerized data.

# 5.0 INDICATORS

## 5.1 Description of NRSA Indicators

### *5.1.1 In Situ Water Quality Measurements*

Measurements for temperature, pH, dissolved oxygen (DO), and conductivity will be taken with a calibrated water quality probe meter or multi-probe sonde at the X-site (center) transect in each river or stream. This information will be used to detect extremes in condition that might indicate impairment.

### *5.1.2 Secchi Disk Transparency*

A Secchi disk is a black and white patterned disk commonly used to measure the clarity of water in visibility distance. It will be used in the boatable systems to determine transparency.

### *5.1.3 Water Chemistry and Associated Measurements*

Water chemistry measurements will be used to determine the acidic conditions and nutrient enrichment, as well as classification of water chemistry type.

### *5.1.4 Chlorophyll-a*

Chlorophyll-a is the pigment that makes plants and algae green. Its measurement is used to determine algal biomass in the water.

### *5.1.5 Sediment Enzymes*

Benthic organisms are in intimate contact with river sediments, and they are influenced by the physical and chemical properties of the sediment. Sediment enzyme activity serves as a functional indicator of key ecosystem processes.

### *5.1.6 Periphyton Assemblage*

Periphyton are diatoms and soft-bodied algae that are attached or otherwise associated with channel substrates. They can contribute to the physical stability of inorganic substrate particles, and provide habitat and structure. Periphyton are useful indicators of environmental condition because they respond rapidly and are sensitive to a number of anthropogenic disturbances, including habitat destruction, contamination by nutrients, metals, herbicides, hydrocarbons, and acidification.

### *5.1.7 Benthic Macroinvertebrate Assemblage*

Benthic macroinvertebrates are bottom-dwelling animals without backbones (“invertebrates”) that are large enough to be seen with the naked eye (“macro”). Examples of macroinvertebrates include: crayfish, snails, clams, aquatic worms, leeches, and the larval and nymph stages of many insects, including dragonflies, mosquitoes, and

mayflies. Populations in the benthic assemblage respond to a wide array of stressors in different ways so that it is often possible to determine the type of stress that has affected a macroinvertebrate assemblage (Klemm et al., 1990). Because many macroinvertebrates have relatively long life cycles of a year or more and are relatively immobile, the structure and function of the macroinvertebrate assemblage is a response to exposure of present or past conditions.

### *5.1.8 Fish Assemblage*

Monitoring of the fish assemblage is an integral component of many water quality management programs. The assessment will measure specific attributes of the overall structure and function of the ichthyofaunal community to evaluate biological integrity and water quality.

### *5.1.9 Physical Habitat Assessment*

The physical habitat assessment of the sampling reach and the riparian zone (the region lying along a bank) will serve three purposes. First, habitat information is essential to the interpretation of what ecological condition is expected to be like in the absence of many types of anthropogenic impacts. Second, the habitat evaluation is a reproducible, quantified estimate of habitat condition, serving as a benchmark against which to compare future habitat changes that might result from anthropogenic activities. Third, the specific selections of habitat information collected aid in the diagnosis of probable causes of ecological degradation in rivers and streams. For example, some of the data collected will be used to calculate relative bed stability (RBS). RBS is an estimate of stream stability that is calculated by comparing the mean sediment size present to the sediment size predicted by channel and slope.

In addition to information collected in the field by the physical habitat assessment, the physical habitat description of each site includes many map-derived variables such as stream order and drainage area. Furthermore, an array of information, including watershed topography and land use, supplements the physical habitat information. Together with water chemistry, the habitat measurements and observations describe the variety of physical and chemical conditions that are necessary to support biological diversity and foster long-term ecosystem stability.

### *5.1.10 Fecal Indicator (Enterococci)*

*Enterococci* are bacteria that are endemic to the guts of warm blooded creatures. These bacteria, by themselves, are not considered harmful to humans but often occur in the presence of potential human pathogens (the definition of an indicator organism). Epidemiological studies of marine and fresh water bathing beaches have established a direct relationship between the density of *enterococci* in water and the occurrence of swimming-associated gastroenteritis. This analysis will not serve as an exact equivalent of a water quality test, since it includes dead organisms as well as living, but it will serve as a surrogate of potential exposure. *Enterococci* samples will be taken from the last transect one meter off the bank.

### *5.1.11 Fish Tissue*

The NRSA fish tissue indicator will provide information on the national distribution of selected persistent, bioaccumulative, and toxic (PBT) chemical residues (e.g., mercury and organochlorine pesticides) in predator fish species from large (non-wadeable) streams and rivers of the conterminous United States. In addition, samples collected from a national statistical subset of NRSA urban sites (approximately 150 sites) located on large (non-wadeable) rivers will be analyzed for pharmaceuticals and personal care product compounds that can persist through the wastewater treatment process. Various studies have been conducted on fish tissue contaminants focusing on different parts of the fish (e.g., whole fish, fillets, livers); however, the NRSA will focus on analysis of fillet tissue because of associated human consumption and health risk implications.

### *5.1.12 Other Indicators / Site Characteristics*

Observations and impressions about the site and its surrounding catchment by field teams will be useful for ecological value assessment, development of associations and stressor indicators, and data verification and validation.

**Table 5-1.** Summary table of indicators

| Indicator | Specs/Location in Sampling Reach |
|---|---|
| In Situ measurements (pH, DO, temperature, conductivity) | One set of measurements taken at midpoint of the river; readings are taken at 0.5 m depth |
| Secchi Disk Transparency | Measurements taken at midpoint of the river; readings are taken at 0.5 m depth |
| Water chemistry (TP, TN [$NH_4$, $NO_3$), basic anions and cations, alkalinity [ANC], DOC, TOC, TSS, conductivity | Collected from a depth of 0.5 m at the midpoint of the river |
| Chlorophyll-a | Collected as part of water chemistry and periphyton samples |
| Sediment enzymes | Collected from 11 locations systematically placed at each site and combined into a single composite sample |
| Periphyton | Collected from 11 locations systematically placed at each site and combined into a single composite sample |
| Benthic macroinvertebrate assemblage (Littoral) | Collected from 11 locations systematically placed at each site and combined into a single composite sample |
| Fish Assemblage | Sampled throughout the sampling reach at specified locations |
| Physical habitat assessment | Measurements collected throughout the sampling reach at specified locations |
| Fecal indicator (*enterococci*) | Collected at the last transect one meter off the bank |
| Fish Tissue | Target species collected throughout the sampling reach |
| Drainage area | Done at desktop, and used in target population selection |
| Characteristics of watershed | Done at desktop using GIS and verified by state agencies |

## 5.2 Water Chemistry

### *5.2.1 Introduction*

Ecological indicators based on river and stream water chemistry information attempt to evaluate stream condition with respect to stressors such as acidic deposition and other types of physical or chemical contamination. Data are collected for a variety of physical and chemical constituents to provide information on the acid-base status of each stream, water clarity, primary productivity, nutrient status, mass balance budgets of constituents, color, temperature regime, and presence and extent of anaerobic conditions.

At each wadeable stream and boatable river site, crews fill one 4L Cubitainer, and a 2L brown plastic bottle. These samples are stored in a cooler packed with resealable plastic bags filled with ice and shipped to the analytical laboratory within 24 hours of collection. Field crews also measure DO, pH, conductivity, and temperature using a multi-parameter water quality meter. Secchi disk depth is only measured at non-wadeable sites. The primary function of the water chemistry information is to determine:

- Acid-base status
- Trophic state (nutrient enrichment)
- Chemical stressors
- Classification of water chemistry type

### *5.2.2 Sampling Design*

The plot design for stream and river sampling is shown in Figure 6. The plot design for water chemistry sampling is based on that used for the National Rivers and Streams Assessment (Kaufmann et al., 1988). At each stream and river, a single sampling site is located at the midpoint of Transect F (the middle transect).

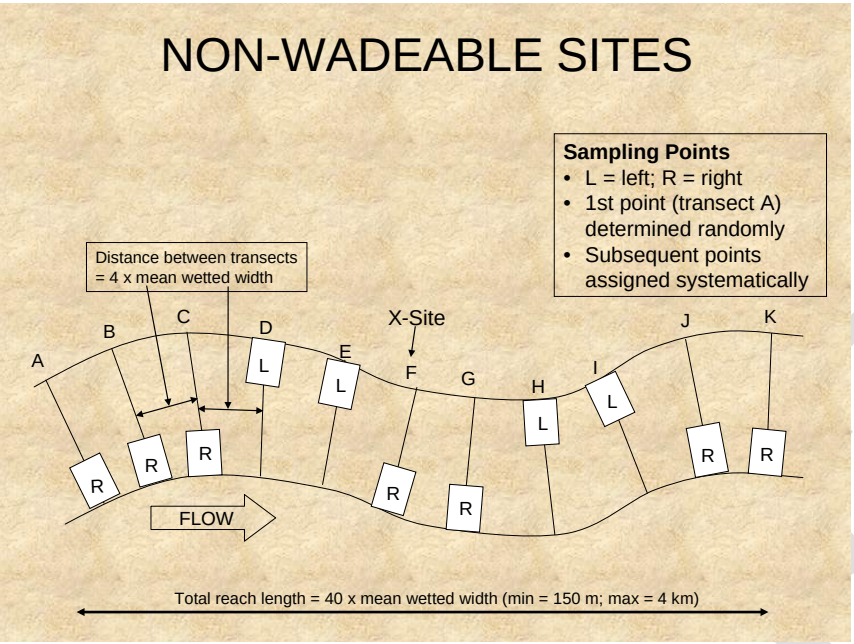


**Figure 6.** Stream and river index sampling design for the water chemistry indicator for non-wadeable sites.


**Figure 7.** Stream and river index sampling design for the water chemistry indicator for wadeable sites.

### *5.2.3 Sampling and Analytical Methodologies*

*Sample Collection:* At wadeable and non-wadeable index sites, a water sample is collected at the midpoint to fill a 4-L cubitainer. A multi-probe sonde is also used at the midpoint to measure DO, pH, temperature, and conductivity.. Secchi disk depths (depths that the disc disappears and reappears) are recorded at the X-site. Detailed procedures for sample collection and handling are described in the field operations manual. Figure 8 presents the process for collecting water chemistry samples and obtaining field measurements.

*Analysis:* Table 5.2-1 summarizes performance requirements for water chemistry and chlorophyll-a analytical methods.. Table 5.2-2 summarizes the analytical methods for the water chemistry indicator. Analytical methods are based on EPA-validated methods, modified for use with aqueous samples of low ionic strength. Modified methods are thoroughly documented in the laboratory methods handbook prepared for the Aquatic Effects Research Program (U.S. EPA, 1987).

### *5.2.4 Quality Assurance Objectives*

Measurement data quality objectives (measurement DQOs or MQOs) are given in Table 19. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.2-3 represent the maximum allowable criteria for statistical control purposes. Method detection limits are monitored over time by repeated measurements of low level standards and calculated using Equation 2-1. For major cations and anions, the required MDLs are approximately equivalent to 1.0 µeq/L (0.5 µeq/L for nitrate). The analytical laboratory may report results in mg/L; these results are converted to µeq/L for interpretation. For total suspended solids determinations, the "detection limit" is defined based on the required sensitivity of the analytical balance.

For precision, the objectives presented in Table 5.2-3 represent the 99% confidence intervals about a single measurement and are thus based on the standard deviation of a set of repeated measurements ($n > 1$). Precision objectives at lower concentrations are equivalent to the corresponding MDL. At higher concentrations, the precision objective is expressed in relative terms, with the 99% confidence interval based on the relative standard deviation (Section 2). Objectives for accuracy are equal to the corresponding precision objective, and are based on the mean value of repeated measurements. Accuracy is generally estimated as net bias or relative net bias (Section 2). For total phosphorus and total nitrogen measurements, accuracy is also determined from analyses of matrix spike samples (also sometimes called fortified samples) as percent recovery (Section 2). Precision and bias are monitored at the point of measurement (field or analytical laboratory) by several types of QC samples described in the Section 5.2.6, and from performance evaluation (PE) samples.

**Table 5.2-1.** Performance requirements for water chemistry and chlorophyll-*a* analytical methods.

| Analyte | Units | Potential Range of Samples[1] | Long-Term MDL Objective[2] | Laboratory Reporting Limit[3] | Transition Value[4] | Precision Objective[5] | Bias Objective[6] |
|---|---|---|---|---|---|---|---|
| Conductivity | µS/cm at 25˚C | 1 to 15,000 | NA | 2.0 | 20 | ± 2 or ±10% | ± 2 or 5% |
| Turbidity | NTU | 0 to 44,000 | 1 | 2.0 | 20 | ± 2 or ±10% | ± 2 or ±10% |
| pH | pH units | 3.7 to 10 | NA | NA | 5.75 and>8.25 | ± 0.08 or ± 0.15 | ± 0.05 or ± 0.10 |
| Acid Neutralizing Capacity (ANC) | µeq/L (20 µeq/L=1 mg as | -300 to +75,000 (-16 to 3,750 mg as $CaCO_3$) | NA | NA | ±50 | ± 5 or ±10% | ± 5 or ±10% |
| Total and Dissolved Organic Carbon (TOC/DOC) | mg C/L | 0.1 to 109 (as DOC) | 0.10 | 0.20 | ≤ 1<br>> 1 | ± 0.10 or ±10% | ± 0.10 or ±10% |
| Ammonia ($NH_3$) | mg N/L | 0 to 17 | 0.01 (0.7 µeq/L) | 0.02 (1.4 µeq/L) | 0.10 | ± 0.01 or ±10% | ± 0.01 or ±10% |
| Nitrate-Nitrite ($NO_3$-$NO_2$) | mg N/L | 0 to 360 (as nitrate) | 0.01 | 0.02 | 0.10 | ± 0.01 or ±10% | ± 0.01 or ±10% |
| Total Nitrogen (TN) | mg/L | 0.1 to 90 | 0.01 | 0.02 | 0.10 | ± 0.01 or ±10% | ± 0.01 or ±10% |
| Total Phosphorus (TP) | µg P/L | 0 to 22,000 | 2 | 4 | 20 | ± 2 or ±10% | ± 2 or ±10% |
| Ortho-phosphate | µgP/L | | 2 | 4 | 20 | ± 2 or ±10% | ± 2 or ±10% |
| Sulfate ($SO_4$) | mg $SO_4$/L | 0 to 5,000 | 0.25 (5 µeq/L) | 0.50 (10 µeq/L) | 2.5 | ± 0.25 or ±10% | ± 0.25 or ±10% |

| Analyte | Units | Potential Range of Samples[1] | Long-Term MDL Objective[2] | Laboratory Reporting Limit[3] | Transition Value[4] | Precision Objective[5] | Bias Objective[6] |
|---|---|---|---|---|---|---|---|
| Chloride (Cl) | mg Cl/L | 0 to 5,000 | 0.10 (3 µeq/L) | 0.20 (6 µeq/L) | 1 | ± 0.10 or ±10% | ± 0.10 or ±10% |
| Nitrate ($NO_3$) | mg N/L | 0 to 360 | 0.01 (1 µeq/L) | 0.02 (4 µeq/L) | 0.1 | ± 0.01 or ±10% | ± 0.01 ±10% |
| Calcium (Ca) | mg Ca/L | 0.04 to 5,000 | 0.05 (2.5 µeq/L) | 0.10 (5 µeq/L) | 0.5 | ± 0.05 or ±10% | ± 0.05 or ±10% |
| Magnesium (Mg) | mg Mg/L | 0.1 to 350 | 0.05 (4 µeq/L) | 0.10 (8 µeq/L) | 0.5 | ± 0.05 or ±10% | ± 0.05 or ±10% |
| Sodium (Na) | mg Na/L | 0.08 to 3,500 | 0.05 (2 µeq/L) | 0.10 (4 µeq/L) | 0.5 | ± 0.05 or ±10% | ± 0.05 or ±10% |
| Potassium (K) | mg K/L | 0.01 to 120 | 0.05 (1 µeq/L) | 0.10 (2 µeq/L) | 0.5 | ± 0.05 or ±10% | ± 0.05 or ±10% |
| Silica ($SiO_2$) | mg $SiO_2$/L | 0.01 to 100 | 0.05 | 0.10 | 0.5 | ± 0.05 or ±10% | ± 0.05 or ±10% |
| Total Suspended Solids (TSS) | mg/L | 0 to 27,000 | 1 | 2 | 10 | ± 1 or ±10% | ± 1 or ±10% |
| True Color | PCU | 0 to 350 | NA | 5 | 50 | ±5 or ±10% | ±5 or ±10% |
| Chlorophyll a | µg/L (in extract) | 0.7 to 11,000 | 1.5 | 3 | 15 | ± 1.5 or ±10% | ± 1.5 or ±10% |

[1] Estimated from samples analyzed at the WED-Corvallis laboratory between 1999 and 2005 for TIME, EMAP-West, and WSA streams from across the U.S.

[2] The long-term method detection limit is determined as a one-sided 99% confidence interval from repeated measurements of a low-level standard across several calibration curves, based on USGS Open File Report 99-193. These represent values that should be achievable by multiple labs analyzing samples over extended periods with comparable (but not necessarily identical) methods.

[3] The minimum reporting limit is the lowest value that need to be quantified (as opposed to just detected), and represents the value of the lowest nonzero calibration standard used. It is set to 2x the long-term detection limit, following USGS Open File Report 99-193 *New Reporting Procedures Based on Long-Term Method Detection Levels and Some Considerations for Interpretations of Water-Quality Data Provided by the U.S. Geological Survey National Water Quality Laboratory.*

[4] Value at which performance objectives for precision and bias switch from absolute (≤ transition value) to relative 9> transition value). Two-tiered approach based on Hunt, D.T.E. and A.L. Wilson. 1986. *The Chemical Analysis of Water: General Principles and Techniques*. 2nd ed.. Royal Society of Chemistry, London, England.

[5] For duplicate samples, precision is estimated as the pooled standard deviation (calculated as the root-mean square) of all samples at the lower concentration range, and as the pooled percent relative standard deviation of all samples at the higher concentration range. For standard samples, precision is estimated as the standard deviation of repeated measurements across batches at the lower concentration range, and as percent relative standard deviation of repeated measurements across batches at the higher concentration range.

[6] Bias (systematic error) is estimated as the difference between the mean measured value and the target value of a performance evaluation and/or internal reference samples at the lower concentration range measured across sample batches, and as the percent difference at the higher concentration range.

**Table 5.2-2:** Analytical methodologies: water chemistry indicator

| Analyte | QA | Expected Range | Summary of Method | References |
|---|---|---|---|---|
| Acid Neutralizing Capacity (ANC) | C | -100 to 5,000 µeq/L | Acidimetric titration to pH ≤ 3.5, with modified Gran plot analysis | EPA 310.1 (modified); U.S. EPA (1987) |
| Carbon, dissolved[a] inorganic (DIC), closed system | N | 0.1 to 50 m g C/L | Sample collected and analyzed without exposure to atmosphere; acid-promoted oxidation to CO2, with detection by infrared spectrophotometry | U.S. EPA (1987) |
| Carbon, dissolved organic (DOC) | C | 0.1 to 30 m g C/L | UV-promoted persulfate oxidation, detection by infrared spectrophotometry. | EPA 415.2, U.S. EPA (1987) |
| Conductivity | C | 1 to 500 µS/cm | Electrolytic (conductance cell and meter) | EPA 120.6, U.S. EPA (1987) |
| *Major Cations (dissolved)* | | | | |
| Calcium<br>Magnesium<br>Sodium<br>Potassium | C<br>C<br>C<br>C | 0.02 to 76 mg/L (1 to 3,800 µeq/L)<br>0.01 to 25 mg/L (1 to 2,000 µeq/L)<br>0.01 to 75 mg/L (0.4 to 3.3 µeq/L)<br>0.01 to 10 mg/L (0.3 to 250 µeq/L) | Atomic absorption spectroscopy (flame) | EPA 200.6, U.S. EPA (1987) |
| Ammonium | N | 0.01 to 5 mg/L (0.5 to 300 µeq/L) | Colorimetric (automated phenate) | EPA 350.7; U.S. EPA (1987) |

| | | | | |
|---|---|---|---|---|
| *Major Anions, dissolved* | | | | |
| Chloride<br>Nitrate<br>Sulfate | C<br>C<br>C | 0.03 to 100 mg/L (1 to 2,800 µeq/L)<br>0.06 to 20 mg/L (0.5 to 350 µeq/L)<br>0.05 to 25 mg/L (1 to 500 µeq/L) | Ion chromatography | EPA 300.6; U.S. EPA (1987) |
| Phosphorus, total | C | 0 to 1000 µg/L | Acid-persulfate digestion with automated colorimetric determination (molybdate blue) | USGS I-4600-78; Skougstad et al. (1979), U.S. EPA (1987) |
| Nitrogen, total | N | 0 to 25,000 µg/L | Alkaline persulfate digestion with determination of nitrate by cadmium reduction and determination of nitrite by automated colorimetry (EDTA/sulfanilimide). | EPA 353.2 (modified); U.S. EPA (1987) |
| Turbidity | N | 1 to 100 Nephelometric Turbidity Units (NTU) | Nephelometric | APHA 214 A., EPA 180.1; U.S. EPA (1987) |
| Total Suspended Solids (TSS) | N | 1 to 200 mg/L | Gravimetric | EPA 160.3; APHA (1989) |

**Table 5.2-3.** Measurement data quality objectives: water chemistry indicator

| Variable or Measurement | Method Detection Limit | Precision and Accuracy | Transition Value[a] | Completeness |
|---|---|---|---|---|
| Oxygen, dissolved | NA | ±0.5 mg/L | NA | 95% |
| Temperature | NA | ±1 ±C | NA | 95% |
| | | | | |
| Acid Neutralizing Capacity | NA | ±5 µeq/L or ±5% | 100 µeq/L | 95% |
| Carbon, dissolved organic | 0.1 mg/L | ±0.1 mg/L or ±10% | 1 mg/L | 95% |
| Conductivity | NA | ±1 µS/cm or ±2% | 50 µS/cm | 95% |
| Major Cations: Calcium Magnesium Sodium Potassium | 0.02 mg/L 0.01 mg/L 0.02 mg/L 0.04 mg/L | ±0.02 mg/L or ±5% ±0.01 mg/L or ±5% ±0.02 mg/L or ±5% ±0.04 mg/L or ±5% | 0.4 mg/L 0.2 mg/L 0.4 mg/L 0.8 mg/L | 95% |
| Ammonium | 0.02 mg/L | ±0.02 mg/L or ±5% | 0.4 mg/L | 95% |
| Major Anions: Chloride Nitrate Sulfate | 0.03 mg/L 0.03 mg/L 0.05 mg/L | ±0.03 mg/L or ±5% ±0.03 mg/L or ±5% ±0.05 mg/L or ±5% | 0.6 m g/L 0.6 m g/L 1 mg/L | 95% |
| Phosphorus, total | 1 µg/L | ±1 µg/L or ±5% | 20 µg/L | 95% |
| Nitrogen, total | 1 µg/L | ±1 µg/L or ±5% | 20 µg/L | 95% |
| Turbidity | NA | ±2 NTU or ±10% | 20 NTU | 95% |
| Total Suspended Solids | 0.1 mg | ±1 mg/L or ±10% | 10 m g/L | 95% |

NA = not applicable

[a] Represents the value above which precision and bias are expressed in relative terms.

### *5.2.5 Quality Control Procedures: Field Operations*

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

Water chemistry field measurements should be measured with a calibrated multiprobe. The DO, pH, and conductivity should be calibrated prior to each sampling event in the field. It is recommended to periodically compare the probe to a DO chemical analysis procedure. Also conduct a quality control check with a different pH and conductivity standard to verify the calibration and periodically evaluate instrument precision. Test the temperature meter against a thermometer that is traceable to the National Institute of Standards (NIST) at least once per sampling season. Field crews should check the calibrated sounding rod and measuring tape attached to the Secchi disk before each sampling event. Field crews should verify that all sample containers are uncontaminated and intact, and that all sample labels are legible and intact. A summary of Field quality control procedures for water chemistry is presented in Table 5.2-4.

Check the label to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the water chemistry sample on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the sample or if conditions occur that may affect sample integrity. Store the sample on wet ice in a cooler. Recheck all forms and labels for completeness and legibility. Additionally, duplicate (replicate) samples will be collected at 10% of sites sampled.

**Table 5.2-4.** Field quality control: Water Chemistry

| Check Description | Frequency | Acceptance Criteria | Corrective Actions |
|---|---|---|---|
| Check calibration of multiprobe | Prior to each sampling day | Specific to instrument | Adjust and recalibrate, redeploy gear |
| Check calibrated sounding rod and measuring tape attached to Secchi disk | Each site | Depth measurements for all sampling points | Obtain best estimate of depth where actual measurement not possible |
| Check integrity of sample containers and | Each site | Clean, intact containers | Obtain replacement supplies |

| | | | |
|---|---|---|---|
| labels | | and labels | |

### *5.2.6 Quality Control Procedures: Laboratory Operations*

#### 5.2.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.2-5. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received. The general schemes for processing stream and river water chemistry samples for analysis is presented in Figure 9. Several additional aliquots are prepared from the bulk water samples. Ideally, all analyses are completed within a few days after processing to allow for review of the results and possible reanalysis of suspect samples within seven days. Critical holding times (Table 5.2-6) for the various analyses are the maximum allowable holding times, based on current EPA and American Public Health Association (APHA) requirements (American Public Health Association, 1989). Analyses of samples after the critical holding time is exceeded will likely not provide representative data.

**Table 5.2-5.** Sample receipt and processing quality control: water chemistry indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| Sample Storage | Store samples in darkness at 4 ºC; Monitor temperature daily | Qualify sample as suspect for all analyses |
| Holding time | Complete processing bulk samples within 48 hours of collection | Qualify samples |
| Aliquot Containers and Preparation | Rinse collection bottles 2 times with stream or river water to be sampled | |
| Filtration | 0.4 µm polycarbonate filters required for all dissolved analytes except DIC (0.45 µm) Rinse filters and filter chamber twice with 50-ml portions of deionized water, followed by a 20-mL portion of sample. Repeat for each filter used on a single sample. Rinse aliquot bottles with two 25 to 50 mL portions of filtered sample before use. | |

| | | |
|---|---|---|
| Preservation | Use ultrapure acids for preservation. Add sufficient acid to adjust to pH < 2. Check pH with indicator paper. Record volume of preservative on container label. Store preserved aliquots in darkness at 4/C until analysis. | |
| Holding Times for preserved aliquots | Holding times range from 3 days to 6 months, based upon current APHA criteria. | Sample results are qualified as being in violation of holding time requirements. |

**Table 5.2-6.** Analyte holding time for various sampling methods

| Analyte | Method | Preservative | Holding time |
|---|---|---|---|
| Total Phosphorus (TP) | USGS I-4600-78 | | |
| Total Nitrogen (TN) | EPA 353.2 | Cool to 4º C | 48 hours |
| Total ammonia-nitrogen ($NH_4$) | ? | | |
| Nitrate ($NO_3$) | EPA 300.6 | | |
| Anions | EPA 300.6 | | |
| Cations | EPA 200.6 | | |
| Total Suspended Solids (TSS) | EPA 160.3 | Cool to 4º C | 7 days |
| Turbidity | EPA 180.1 | Cool to 4º C | 4 hours |
| Acid Neutralizing Capacity (ANC, alkalinity) | EPA 310.1 | Cool to 4º C | 14 days |
| Dissolved Organic Carbon (DOC) | EPA 415.2 | | |

#### 5.2.6.2 Analysis of Samples

QC protocols are an integral part of all analytical procedures to ensure that the results are reliable and the analytical stage of the measurement system is maintained in a state of statistical control. Most of the QC procedures described here are detailed in the references for specific methods. However, modifications to the procedures and acceptance criteria described in this QAPP supersede those presented in the methods references. Information regarding QC sample requirements and corrective actions are summarized in Table 5.2-7. Figure 9 illustrates the general scheme for analysis of a batch of water chemistry samples, including associated QC samples.

### *5.2.7 Data Reporting, Review, and Management*

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.2-8. Data reporting units and significant figures are given in Table 5.2-9. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members.

**FIELD MEASUREMENT PROCESS: WATER CHEMISTRY INDICATOR**

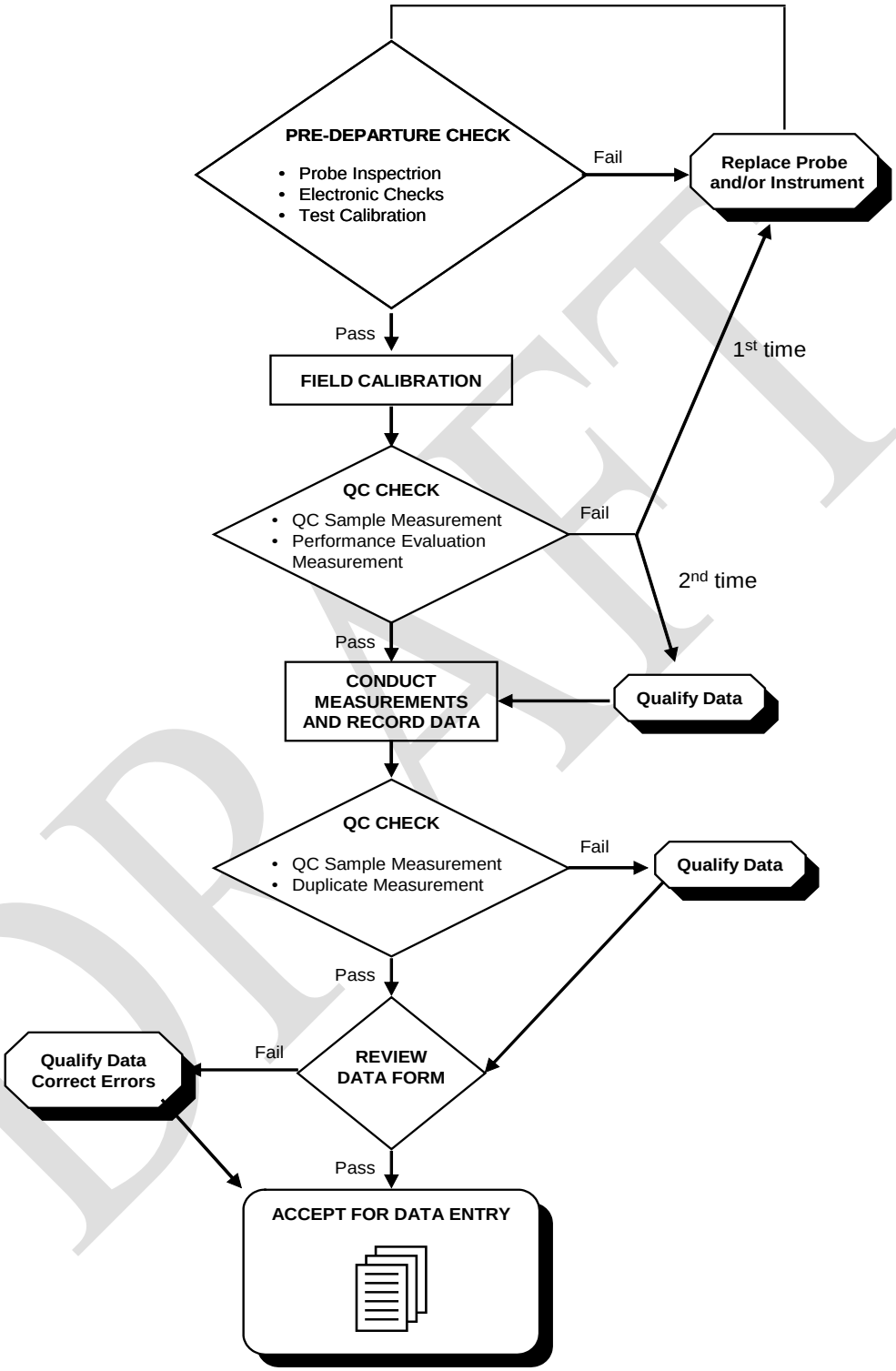

**Figure 8:** Field Measurement process for water chemistry samples.

**Table 5.2-7.** Laboratory quality control samples: water chemistry indicator

| QC Sample Type (Analytes), and Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Laboratory Blank: (all analyses total suspended solids[TSS]) Reagent Blank: (DOC, Al [total, monomeric, and organic monomeric], ANC, NH4+, SiO2) | Once per batch prior to sample analysis | Control limits < ±MDL | Prepare and analyze new blank. Determine and correct problem (e.g., reagent contamination, instrument calibration, or contamination introduced during filtration) before proceeding with any sample analyses. Reestablish statistical control by analyzing three blank samples. |
| Filtration Blank: (All dissolved analytes, excluding syringe samples) ASTM Type II reagent water processed through filtration unit. | Prepare 1/week and archive | Measured concentrations < MDL | Measure archived samples if review of other laboratory blank information suggest source of contamination is sample processing. |
| Detection Limit Quality Control Check Sample (QCCS): (All analyses except true color, turbidity, and TSS) Prepared so concentration is approximately 4-6 times the required MDL. | Once per batch | Control limits < ±MDL | Confirm achieved MDL by repeated analysis of appropriate standard solution. Evaluate affected samples for possible re-analysis. |
| Calibration QCCS: For turbidity, QCCS is prepared at one level for routine analyses (USEPA 1987). Additional QCCS are prepared as needed for samples having estimated turbidities >20 NTU. For TSS determinations, QCCS is a standard weight having mass representative of samples. | Before and after sampl | Control limits < precision objective: Mean value < bias objective | Repeat QCCS analysis. Recalibrate and analyze QCCS. Reanalyze all routine samples (including PE and field replicate samples) analyzed since the last acceptable QCCS measurement. |

| | | | |
|---|---|---|---|
| | e an al ys es | | |
| Internal Reference Sample: (Suggested when available for a particular analyte) | One an al ysi s in a mi ni m u m of fiv e se pa rat e ba tc he s | Control limits < precision objective. Mean value < bias objective | Analyze standard in next batch to confirm suspected imprecision or bias. Evaluate calibration and QCCS solutions and standards for contamination and preparation error. Correct before any further analyses of routine samples are conducted. Reestablish control by three successive reference standard measurements which are acceptable. Qualify all sample batches analyzed since the last acceptable reference standard measurement for possible reanalysis. |
| Laboratory Replicate Sample: (All analyses) For closed system analyses, a replicate sample represents a second injection of sample from the sealed syringe. | One per ba tc h | Control limits < precision objective | If results are below MDL: Prepare and analyze split from different sample (volume permitting). Review precision of QCCS measurements for batch. Check preparation of split sample. Qualify all samples in batch for possible reanalysis. |
| Matrix spike samples: (Only prepared when samples with potential for matrix interferences are encountered) | One per ba tc h | Control limits for recovery cannot exceed 100±20% | Select two additional samples and prepare fortified subsamples. Reanalyze all suspected samples in batch by the method of standard additions. Prepare three subsamples (unfortified, fortified with solution approximately equal to the endogenous concentration, and fortified with solution |

| | | | |
|---|---|---|---|
| | | | approximately twice the endogenous concentration. |

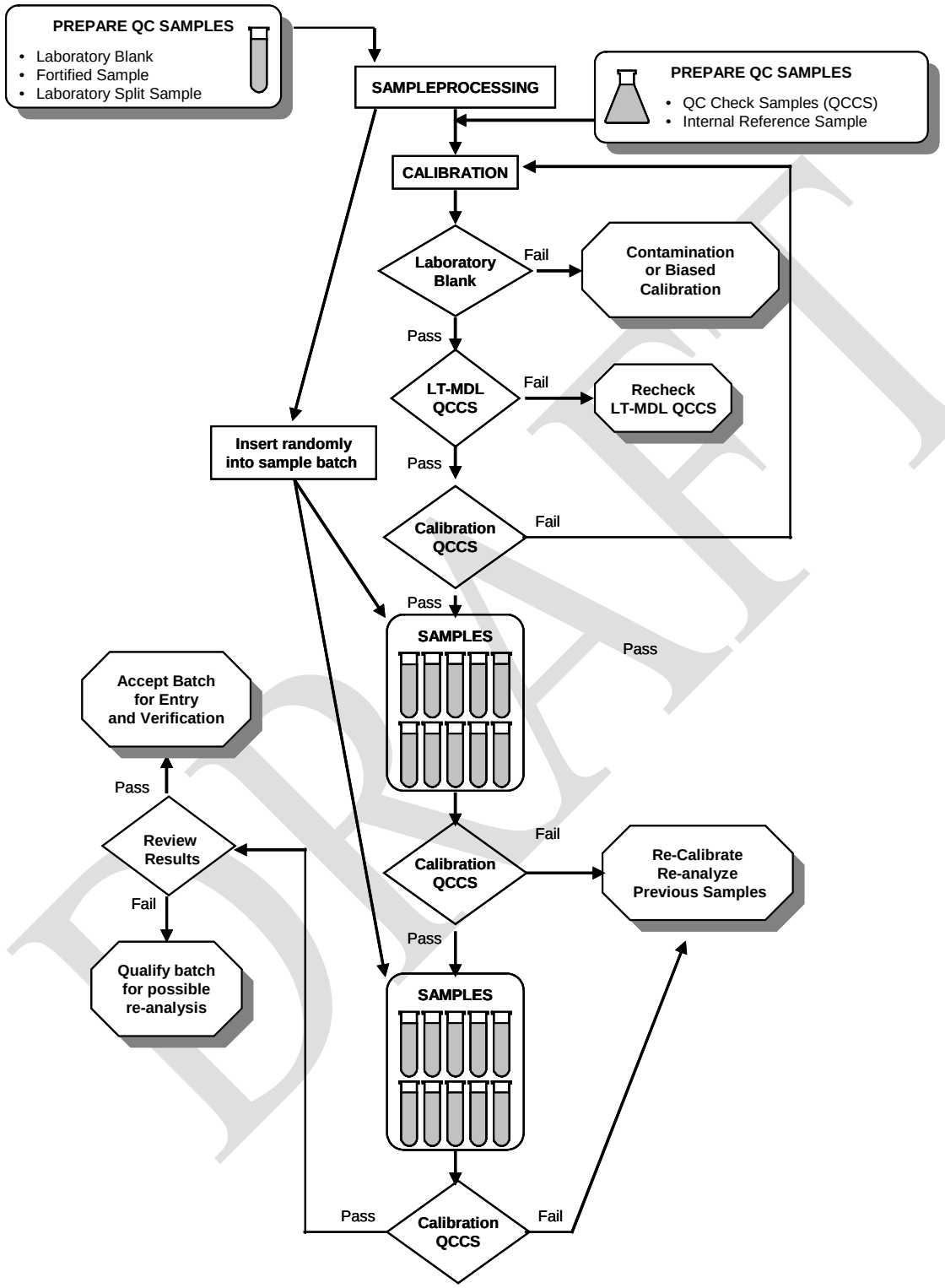


**Figure 9.** Analysis activities for water chemistry samples.

**Table 5.2-8.** Data review, verification, and validation quality control: water chemistry indicator

| Activity or Procedure | Requirements and Corrective Action |
|---|---|
| Range checks, summary statistics, and/or exploratory data analysis (e.g., box and whisker plots) | Correct reporting errors or qualify as suspect or invalid. |
| Review holding times | Qualify value for additional review |
| Ion balance: Calculate percent ion balance difference (%IBD) using data from cations, anions, and ANC. | If total ionic strength ≤100 µeq/L, %IBD ≤±25%. If total ionic strength >100 µeq/L, %IBD ≤±10%. Determine which analytes, if any, are the largest contributors to the ion imbalance. Review suspect analytes for analytical error and reanalyze. If analytical error is not indicated, qualify sample to attribute imbalance to unmeasured ions. Reanalysis is not required. Flag= %IBD outside acceptance criteria due to unmeasured ions |
| Conductivity check: Compare measured conductivity of each sample to a calculated conductivity based on the equivalent conductances of major ions in solution (Hillman et al., 1987). | If measured conductivity ≤ 25 µS/cm, ([measured ! calculated] ÷ measured) ≤ ±25%. If measured conductivity > 25 µS/cm, ([measured ! calculated] ÷ measured) ≤ ±15%. Determine which analytes, if any, are the largest contributors to the difference between calculated and measured conductivity. Review suspect analytes for analytical error and reanalyze. If analytical error is not indicated, qualify sample to attribute conductivity difference to unmeasured ions. Reanalysis is not required. |
| Aluminum check: Compare results for organic monomeric aluminum, total monomeric aluminum, and total dissolved aluminum. | [organic monomeric] < [total monomeric] < [total dissolved]. Review suspect measurement(s) to confirm if analytical error is responsible for inconsistency. |
| ANC check: Calculate ANC based on pH and DIC. Compare to measured ANC | Review suspect measurements for samples with results outside of acceptance criteria. Determine if analytical error or non-carbonate alkalinity are responsible for lack of agreement. |
| Review data from QA samples (laboratory PE samples, and interlaboratory comparison samples) | Compare with results from other years to determine comparability. Determine impact and possible limitations on overall usability of data |

**Table 5.2-9.** Data reporting criteria: water chemistry indicator

| Measurement | Units | Significant Figures | Maximum Decimal Places |
|---|---|---|---|
| Dissolved Oxygen | mg/L | 2 | 1 |
| Temperature | ºC | 2 | 1 |

| | | | |
|---|---|---|---|
| pH | pH units | 3 | 2 |
| Carbon, dissolved organic | mg/L | 3 | 1 |
| Acid neutralizing capacity | µeq/L | 3 | 1 |
| Conductivity | µS/cm at 25 /C | 3 | 1 |
| Calcium, magnesium, sodium, potassium, ammonium, chloride, nitrate, and sulfate | µeq/L | 3 | 1 |
| Total phosphorus and total nitrogen | µg/L | 3 | 0 |
| Turbidity | NTU | 3 | 0 |
| Total suspended solids | mg/L | 3 | 1 |

The ion balance for each sample is computed using the results for major cations, anions, and the measured acid neutralizing capacity. The percent ion difference (%IBD) for a sample is calculated as:

$$\%IBD = \frac{(\Sigma\ cations - \Sigma\ anions) - ANC}{ANC + \Sigma\ anions + \Sigma\ cations + 2[H^+]}$$

where ANC is the acid neutralization capacity, cations are the concentrations of calcium, magnesium, sodium, potassium, and ammonium, converted from mg/L to µeq/L, anions are chloride, nitrate, and sulfate (converted from mg/L to µeq/L), and $H^+$ is the hydrogen ion concentration calculated from the antilog of the sample pH. Factors to convert major ions from mg/L to µeq/L are presented in Table 5.2-10. For the conductivity check, equivalent conductivities for major ions are presented in Table 5.2-11.

**Table 5.2-10.** Constants for converting major ion concentrations from mg/L to µeq/L

| Analyte | Conversion from mg/L to µeq/La |
|---|---|
| Calcium | 49.9 |
| Magnesium | 82.3 |
| Potassium | 25.6 |
| Sodium | 43.5 |
| Ammonium | 55.4 |
| Chloride | 28.2 |
| Nitrate | 16.1 |
| Sulfate | 20.8 |

[a] Measured values are multiplied by the conversion factor.

**Table 5.2-11.** Factors to calculate equivalent conductivities of major ions[a]

| Ion | Equivalent | Ion | Equivalent |
|---|---|---|---|

| | Conductance per mg/L (µS/cm at 25 /C) | | Conductance per mg/L (µS/cm at 25 /C) |
|---|---|---|---|
| Calcium | 2.60 | Nitrate | 1.15 |
| Magnesium | 3.82 | Sulfate | 1.54 |
| Potassium | 1.84 | Hydrogen | 3.5 × 105 b |
| Sodium | 2.13 | Hydroxide | 1.92 × 105 *b* |
| Ammonium | 4.13 | Bicarbonate | 0.715 |
| Chloride | 2.14 | Carbonate | 2.82 |

[a] From Hillman et al. (1987).

[b] Specific conductance per mole/L, rather than per mg/L.

## 5.3 Chlorophyll-*a* Indicator

### 5.3.1 Introduction

Data are collected for chlorophyll-*a* to provide information on the algal loading and gross biomass of blue-greens and other algae within each stream and river.

### 5.3.2 Sampling Design

The samples are collected at the index site located at the midpoint of the center transect of the reach (transect F) on wadeable and non-wadeable sites. The plot design for sampling locations is shown in Figure 6.

### 5.3.3 Sampling and Analytical Methods

*Sample Collection:* At the index site, collect a 2-L water sample from the surface using the Nalgene beaker and transfer sample immediately to the 2-L brown bottle. The sample should be preserved immediately on ice and placed in a cooler away from direct light. After returning to shore, the sample is filtered in subdued light to minimize degradation. The filter is then stored in a centrifuge tube on ice before being shipped to the laboratory for chlorophyll-*a* analysis. Detailed procedures for sample collection and processing are described in the Field Operations Manual.

*Analysis:* A performance-based methods approach is being utilized for chlorophyll-*a* analysis that defines a set of laboratory method performance requirements for data quality. Following this approach, participating laboratories may choose which analytical method they will use to determine chlorophyll-a concentration as long as they are able to achieve the performance requirements as listed in Table 5.2-1.

### 5.3.4 Quality Assurance Objectives

MQOs are given in Table 5.2-1. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.2-1 represent the maximum allowable criteria for statistical control purposes. LT-MDLs are monitored over time by repeated measurements of low level standards and calculated using Equation 1a.

For precision, the objectives presented in Table 5.2-1 represent the 99% confidence intervals about a single measurement and are thus based on the standard deviation of a set of repeated measurements (n > 1). Precision objectives at lower concentrations are equivalent to the corresponding LRL. At higher concentrations, the precision objective is expressed in relative terms, with the 99% confidence interval based on the relative standard deviation (Section 2). Objectives for accuracy are equal to the corresponding precision objective, and are based on the mean value of repeated measurements. Accuracy is generally estimated as net bias or relative net bias (Section 2). Precision and bias are monitored at the point of measurement (field or analytical laboratory) by several types of QC samples described in Table 5.2-7, where applicable, and from performance evaluation (PE) samples.

### 5.3.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

Chlorophyll can degrade rapidly when exposed to bright light. It is important to keep the sample on ice and in a dark place (cooler) until it can be filtered. If possible, prepare the sample in subdued light (or shade) by filtering as quickly as possible to minimize degradation. If the sample filter clogs and the entire sample in the filter chamber cannot be filtered, discard the filter and prepare a new sample, using a smaller volume.

Check the label to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the chlorophyll-*a* sample on the Sample Collection Form. Also record the volume of sample filtered on the Sample Collection Form. Verify that the volume recorded on the label matches the volume recorded on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the sample or if conditions occur that may affect sample integrity. Store the filter sample in a 50-mL centrifuge tube (or other suitable container) wrapped in aluminum foil and freeze using dry ice or a portable freezer. Recheck all forms and labels for completeness and legibility. Additionally, duplicate (replicate) samples will be collected at 10% of sites sampled. A summary of field quality control procedures for the chlorophyll-a sample is presented in Table 5.3-1.

**Table 5.3-1.** Sample collection and field processing quality control: chlorophyll-a indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Sample Storage (field) | Store sample on wet ice and in a dark place (cooler) | Discard and recollect sample |
| Sample Processing (field) | Filter the sample quickly in a shaded area to minimize degradation | Qualify samples |
| Filtration (done in field) | Whatman GF/F (or equivalent) glass fiber filter. Filtration pressure should not exceed 7 psi to avoid rupture of fragile algal cells. | Discard and refilter |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |
| Holding time | Frozen filter must be shipped on wet ice immediately | Qualify samples |

### 5.3.6 Quality Control Procedures: Laboratory Operations

#### 5.3.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.3-2. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received.

**Table 5.3-2.** Sample receipt and processing quality control: chlorophyll-*a* indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| | | |
| Sample Storage | Store samples in darkness and frozen (-20 °C) Monitor temperature daily | Qualify sample as suspect for all analyses |

### 5.3.6.2 Analysis of Samples

QC protocols are an integral part of all analytical procedures to ensure that the results are reliable and the analytical stage of the measurement system is maintained in a state of statistical control. Most of the QC procedures described here are detailed in the references for specific methods. However, modifications to the procedures and acceptance criteria described in this QAPP supersede those presented in the methods references. QC activities associated with sample analysis are presented in Table 5.3-3.

**Table 5.3-3.** Sample analysis quality control: chlorophyll-*a* indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| | | |
| | | |
| | | |

### 5.3.7 Data Reporting, Review, and Management

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.3-4. Data reporting units and significant figures are given in Table 5.3-5. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA. The electronic data files are transferred to the NRSA IM Coordinator at WED-Corvallis for entry into a centralized data base. A hard copy output of all files will also be sent to the NRSA IM Coordinator.

**Table 5.3-4.** Data review, verification, and validation quality control: chlorophyll-*a* indicator

| Activity or Procedure | Requirements and Corrective Action |
|---|---|
| Range checks, summary statistics, and/or exploratory data analysis (e.g., box and whisker plots) | Correct reporting errors or qualify as suspect or invalid |
| Review data from QA samples (e.g., laboratory PE samples or other standards or replicates) | Determine impact and possible limitations on overall usability of data |

**Table 5.3-5.** Data reporting criteria: chlorophyll-*a* indicator

| Measurement | Units | No. Significant Figures | Maximum No. Decimal Places |
|---|---|---|---|
| Chlorophyll-*a* | µg/L | 2 | 1 |

## 5.4 Sediment Enzymes Indicator

### 5.4.1 Introduction

Benthic organisms are in intimate contact with river sediments, and they are influenced by the physical and chemical properties of the sediment. Sediment enzyme activity serves as a functional indicator of key ecosystem processes. Sediment samples are collected, preserved and analyzed to determine extracellular enzyme activity using the Bio-tek microplate reader of fluorescence/luminescence.

### 5.4.2 Sampling Design

The samples are collected at the 11 sampling stations at each site and combined, resulting in a single 500 mL composite sample per site. The transect and plot design for sampling locations is shown in Figure 6.

### 5.4.3 Sampling and Analytical Methods

*Sample Collection:* Collect sediment samples at the 11 transect sampling stations at each site and combine all subsamples at a site, resulting in a single 500 mL composite sample per site. Collect fine surface sediments (top 5 cm) using a stainless steel spoon or dredge. Store the samples on wet ice in the field. If not shipped immediately, samples may be stored in a refrigerator for no more than 2 weeks until shipment to the analytical laboratory for processing. Samples will be analyzed for available DIN, NH4, DIP, TP, TN, total carbon (TC), and enzyme activity. Detailed procedures for sample collection and processing are described in the Field Operations Manual.

*Analysis:* Sediment samples are collected in clean ziplock bags and frozen until analysis. The subsamples are weighed (0.5-2.0g wet weight) into 125mL Nalgene bottles and either refrozen until analysis, or used immediately. Seventy-five (75) ml acetate buffer is added to sample, homogenized, and then quantitatively transferred to a 300 ml sterile wide mouth glass jar. An additional 125 ml of buffer is added, and re-homogenized if necessary. Prepared samples are stored in the refrigerator, and stirred with stir bar during sample pipetting. Samples are run (or diluted and run) on the Bio-tek fluorescence detector. Detailed procedures are contained in the laboratory operations manual and cited references.

### 5.4.4 Quality Assurance Objectives

MQOs are given in Table 5.4-1. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.4-1 represent the maximum allowable criteria for statistical control purposes. LT-MDLs are monitored over time by repeated measurements of low level standards and calculated using Equation 1a.

**Table 5.4-1.** Measurement data quality objectives: sediment enzymes indicator

| Variable or | Method | Precision and | Transition | Completeness |
|---|---|---|---|---|

| Measurement | Detection Limit | Accuracy | Value[a] | |
|---|---|---|---|---|
| DIN | | | | |
| NH4 | | | | |
| DIP | | | | |
| TP | | | | |
| TN | | | | |
| total carbon (TC) | | | | |
| enzyme activity | | | | |
| | | | | |
| | | | | |
| | | | | |

NA = not applicable

[a] Represents the value above which precision and bias are expressed in relative terms.

### 5.4.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

It is important to keep the individual sediment subsamples on wet ice and in a dark place (cooler) as each subsequent subsample is collected. After the subsamples are composited, the composite sample is stored on wet ice and in a dark place (cooler in field; refrigerator in lab). The composited samples must be shipped to the analytical laboratory within 2 weeks of collection.

Check the sample label to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the sediment sample on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the sample or if conditions occur that may affect sample integrity. Recheck all forms and labels for completeness and legibility. Additionally, duplicate (replicate) samples will be collected at 10% of sites sampled. A summary of field quality control procedures for sediment enzyme samples is presented in Table 5.4-2.

**Table 5.4-2.** Sample collection and field processing quality control: sediment enzymes indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Sample Storage (field) | Store sediment samples on wet ice and in a dark place (cooler) | Discard and recollect sample |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |
| Holding time | Refrigerated samples must be shipped on wet ice within 2 weeks of collection | Qualify samples |
| Sample Storage (lab) | Sediment samples are collected in clean ziplock bags and frozen until analysis. | Qualify sample as suspect for all analyses |

### 5.4.6 Quality Control Procedures: Laboratory Operations

#### 5.4.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.4-3. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received.

**Table 5.4-3.** Sample receipt and processing quality control: sediment enzymes indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| | | |
| | | |
| | | |
| | | |

#### 5.4.6.2 Analysis of Samples

QC protocols are an integral part of all analytical procedures to ensure that the results are reliable and the analytical stage of the measurement system is maintained in a state of statistical control. Most of the QC procedures described here are detailed in the

references for specific methods. However, modifications to the procedures and acceptance criteria described in this QAPP supersede those presented in the methods references. Replicate lab samples should be analyzed on at least 10% of total number of samples analyzed. Replicate lab samples should agree within 20-30% of each determination. QC activities associated with sample receipt and processing are presented in Table 5.4-4. (*There is very little QA/QC info in the Lab SOP; need more info for this section*)

**Table 5.4-4.** Sample analysis quality control: sediment enzymes indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| | | |
| | | |
| | | |
| | | |
| | | |

### 5.4.7 Data Reporting, Review, and Management

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.4-5. Data reporting units and significant figures are given in Table 5.4-6. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA. The electronic data files are transferred to the NRSA IM Coordinator at WED-Corvallis for entry into a centralized data base. A hard copy output of all files will also be sent to the NRSA IM Coordinator.

**Table 5.4-5.** Data review, verification, and validation quality control: sediment enzymes indicator

| Activity or Procedure | Requirements and Corrective Action |
|---|---|
| Range checks, summary statistics, and/or exploratory data analysis (e.g., box and whisker plots) | Correct reporting errors or qualify as suspect or invalid |
| Review data from QA samples (e.g., laboratory PE samples or other standards or replicates) | Determine impact and possible limitations on overall usability of data |

**Table 5.4-6.** Data reporting criteria: sediment enzymes indicator

| Measurement | Units | No. Significant Figures | Maximum No. Decimal Places |
|---|---|---|---|

| | | | |
|---|---|---|---|
| DIN | | | |
| NH4 | | | |
| DIP | | | |
| TP | | | |
| TN | | | |
| total carbon (TC) | | | |
| enzyme activity | | | |

## 5.5 Periphyton

### 5.5.1 Introduction

Periphyton are diatoms and soft-bodied algae that are attached or otherwise associated with channel substrates. They can contribute to the physical stability of inorganic substrate particles, and provide habitat and structure. Periphyton are useful indicators of environmental condition because they respond rapidly and are sensitive to a number of anthropogenic disturbances, including habitat destruction, contamination by nutrients, metals, herbicides, hydrocarbons, and acidification.

### 5.5.2 Sampling Design

The samples are collected at the 11 sampling stations at each site and combined, resulting in a single 500 mL composite sample per site. Four individual samples are prepared from this composite sample. The transect and plot design for sampling locations is shown in Figure 6.

### 5.5.3 Sampling and Analytical Methodologies

*Sample Collection:* At the each transect within the littoral zone, crews collect periphyton samples from coarse substrate. A 12cm delimiter is used to define the sampling area on the substrate. An aspirator is used if no coarse substrate is available. The sample is a composite from each of the 11 transects throughout the reach. In the post-sampling activities, periphyton composite samples will be separated for a 50 ml community sample, a filtered ash free dry mass sample, a filtered chlorophyll-a sample and a 50 ml acid phosphotase activity sample.

*Analysis:* Community identification samples are preserved, processed, enumerated, and organisms identified to the lowest possible taxonomic level (generally genus, see Laboratory Methods Manual) using specified standard keys and references. Processing and archival methods are based on USGS NAWQA methods (Charles et al. 2003). Detailed procedures are contained in the laboratory methods manual and cited references. There is no maximum holding time associated with preserved periphyton samples. Chlorophyll-a samples will be filtered on a Whatman GF/F 0.7µm filter, frozen in the filed and shipped to the Dynamac lab. The sample analysis and QC will follow that previously described for water column chlorophyll-a in section 5.2. Acid Phosphatase Activity (APA) samples will be frozen in the field and shipped on ice to the analysis lab in Duluth, MN. Ash free dry mass samples will be filtered in the field, and filters shipped to the analytical lab.

### 5.5.4 Quality Assurance Objectives

MQOs are given in Table 5.5-1. General requirements for comparability and representativeness are addressed in Section 2. Precision is calculated as percent efficiency, estimated from independent identifications of organisms in randomly selected samples. The MQO for accuracy is evaluated by having individual specimens representative of selected taxa identified by recognized experts.

**Table 5.5-1.** Measurement data quality objectives: phytoplankton indicator

| Variable or Measurement | Precision | Accuracy | Completeness |
|---|---|---|---|
| Enumeration | 85% | 90%[a] | 99% |
| Identification | 85% | 90%[a] | 99% |

[a] Taxonomic accuracy, as calculated using Equation 9 in Section 2.

### 5.5.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

It is important to keep the individual periphyton subsamples on wet ice and in a dark place (cooler) as each subsequent subsample is collected. After the 500-mL bottle has been filled, the composite sample is processed (filtered or preserved) in the field. The sample must be thoroughly mixed before processing to ensure that the sample material is evenly distributed throughout the composite. The crews must be careful to use the appropriate filter or preservative for each type of sample prepared from the composite.

The sample labels should be checked to ensure that all written information is complete and legible, and that the label has been completely covered with clear packing tape. It should be verified that the bar code assigned to the periphyton samples is recorded correctly on the Sample Collection Form. The presence of preservative in the sample should be noted on the Sample Collection Form to assure the integrity of the sample. A flag code should be recorded and comments provided on the Sample Collection Form to denote any problems encountered in collecting the sample or the presence of any conditions that may affect sample integrity. Recheck all forms and labels for completeness and legibility. Additionally, duplicate (repeat) samples will be collected at 10% of lakes sampled. A summary of Field quality control procedures for periphyton samples is presented in Table 5.5-2.

**Table 5.5-2.** Sample collection and field processing quality control: periphyton indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Sample Storage (field) | Store samples on wet ice and in a dark place (cooler) | Discard and recollect sample |

| | | |
|---|---|---|
| Homogenize composite | Thoroughly mix samples before processing to ensure that the sample material is evenly distributed throughout the composite. | Discard and recollect sample |
| Preparing samples | Use the appropriate filter or preservative for each type of sample prepared from the composite. | Discard and prepare a replacement subsample from the composite |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |
| Holding times | The frozen chlorophyll and AFDM filters are shipped immediately on wet ice. The APA sample may be held frozen and shipped on wet ice within 2 weeks of collection. The ID sample preserved with Lugol's solution is held in a refrigerator and must be shipped on wet ice within 2 weeks of collection. | Qualify samples |

### 5.5.6 Quality Control Procedures: Laboratory Operations

#### 5.5.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.5-3. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received.

**Table 5.5-3.** Sample receipt and processing quality control: periphyton indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| Sample Storage | | Qualify sample as suspect for all analyses |
| Holding time | | Qualify samples |
| Filtration | | Qualify samples |
| Preservation | | Qualify samples |

#### 5.5.6.2 Analysis of Samples

It is critical that prior to taking a small portion of the subsample, the sample be thoroughly mixed and macro or visible forms are evenly dispersed.

### 5.5.7 Data Management, Review, and Validation

The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA project. The electronic data files are transferred to the Rivers and Streams Survey IM Coordinator at WED-Corvallis for entry into a centralized data base. A hard copy output of all files will also be sent to the Rivers and Streams Survey IM Coordinator.

Sample residuals, vials, and slides are archived by each laboratory until the EPA Project Leader has authorized, in writing, the disposition of samples. All raw data (including field data forms and bench data recording sheets) are retained permanently in an organized fashion by the Indicator Lead in accordance with EPA records management policies.

## 5.6 Benthic Macroinvertebrates

### 5.6.1 Introduction

The benthic macroinvertebrate assemblage found in sediments and on substrates of streams and rivers reflect an important aspect of the biological condition of the stream or river. The response of benthic communities to various stressors can often be used to determine the type of stressor and to monitor trends (Klemm et al., 1990). The overall objectives of the benthic macroinvertebrate indicators are to detect stresses on community structure in National rivers and streams and to assess and monitor the relative severity of those stresses. The benthic macroinvertebrate indicator procedures are based on various recent bioassessment literature (Barbour et al. 1999, Hawkins et al. 2000, Peck et al. 2003).

### 5.6.2 Sampling Design

Benthic macroinvertebrates are collected at randomly selected sampling locations on the 11 cross-sectional transects established along the stream reach. A composite sample is collected from a multi-habitat approach and consists of sampling pools, riffles, runs, and glides. See field manual for more details.

### 5.6.3 Sampling and Analytical Methodologies

*Sample Collection:* Benthic macroinvertebrate composite samples are collected using a D-frame net with 500 µm mesh openings. The samples are taken from the randomly selected sampling stations at the 11 transects equally distributed along the targeted reach. Benthic macroinvertebrates are collected from an approximately 1 $ft^2$ area in wadeable systems and from 1 linear meter in non-wadeable systems. Samples are field-processed to remove large detritus (rinsed and inspected for organisms) and preserved in ethanol. Detailed sampling and processing procedures are described in the

field operations manual. A condensed description of key elements of the field activities is provided for easy reference onsite.

*Analysis:* Preserved composite samples are sorted, enumerated, and invertebrates identified to the genus level (see Attachment 6 of the Laboratory Methods Manual) using specified standard keys and references. Processing and archival methods are based on standard practices. Detailed procedures are contained in the laboratory methods manual and cited references. There is no maximum holding time associated with preserved benthic macroinvertebrate samples. Five hundred benthic organism count is the target number to match the EMAP West protocol. A 10% external check is standard QA for EMAP West. For operational purposes of the NRSA, laboratory sample processing should be completed by March 2010. Table 5.6-1 summarizes field and analytical methods for the benthic macroinvertebrates indicator.

**Table 5.6.1.** Field and laboratory methods: benthic indicator

| Variable or Measurement | QA | Expected Range/Units | Summary of Method | References |
|---|---|---|---|---|
| Sample Collection | C | NA | D-frame kick net (500 µm mesh) used to collect organisms, which are composited from 11 transects | Barbour et al. 1999, Peck et al. 2003, WSA Field Operation Manual 2004 |
| Sorting and Enumeration | C | 0 to 500 organisms | Random systematic selection of grids with target of 500 organisms from sample | W SA Benthic Laboratory Methods 2004 |
| Identification | C | genus | Specified keys and references | |

C = critical, N = non-critical quality assurance classification.

### 5.6.4 Quality Assurance Objectives

Measurement quality objectives (MQOs) are given in Table 5.8-2. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 8 represents the maximum allowable criteria for statistical control purposes. Precision is calculated as percent efficiency, estimated from examination of randomly selected sample residuals by a second analyst and independent identifications of organisms in randomly selected samples. The MQO for picking accuracy is estimated from examinations (repicks) of randomly selected residues by experienced taxonomists.

**Table 5.6.2.** Measurement data quality objectives: benthic indicator

| Variable or Measurement | Precision | Accuracy | Completeness |
|---|---|---|---|
| Sort and Pick | 95% | 90% | 99% |
| Identification | 85% | 90%a | 99% |

NA = not applicable
[a]Taxonomic accuracy, as calculated using Equation 10 in Section 2.

The completeness objectives are established for each measurement *per site type* (e.g., probability sites, revisit sites, etc.). Failure to achieve the minimum requirements for a particular site type results in regional population estimates having wider confidence intervals. Failure to achieve requirements for repeat and annual revisit samples reduces the precision of estimates of index period and annual variance components, and may impact the representativeness of these estimates because of possible bias in the set of measurements obtained.

### 5.6.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

It is important to keep the individual benthic macroinvertebrate subsamples wet while in the sieve bucket as each subsequent subsample is collected. It is recommended that teams carry a sample bottle containing a small amount of ethanol with them to enable them to immediately preserve larger predaceous invertebrates such as helgramites and water beetles. Doing so will help reduce the chance that other specimens will be consumed or damaged prior to the end of the field day. Once the composite sample from all stations is sieved and reduced in volume, store in a 1-liter jar and preserve with 95% ethanol. Do not fill jars more than 1/3 full of material to reduce the chance of organisms being damaged or crushed during transport. The composite sample is stored in a cool, dark place until it is shipped to the analytical laboratory.

Check the sample label to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the benthic sample on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the sample or if conditions occur that may affect sample integrity. Recheck all forms and labels for completeness and legibility. Additionally, duplicate (replicate) samples will be collected at 10% of sites sampled. Specific quality control measures are listed in Table 5.6-3 for field operations.

**Table 5.6-3.** Sample collection and field processing quality control: benthic indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Sample Collection | Keep the individual benthic macroinvertebrate subsamples wet while in the sieve bucket as each subsequent subsample is collected. | |
| Sample Collection | Carry a small amount of ethanol to immediately preserve larger predaceous invertebrates to reduce the chance that other specimens will be consumed or damaged. | Qualify samples |
| Sample Processing (field) | Preserve with 95% ethanol. Fill jars1/3 full of material to reduce the chance of organisms being damaged. | |

| | | |
|---|---|---|
| Sample Storage (field) | Store benthic samples in a cool, dark place until shipment to analytical lab | Discard and recollect sample |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |
| Holding time | Preserved samples can be stored indefinitely; periodically check jars and change the ethanol if sample material appears to be degrading. | Qualify samples |

### 5.6.6 Quality Control Procedures: Laboratory Operations

#### 5.6.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.6-4. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received.

**Table 5.6-4.** Sample receipt and processing quality control: benthic macroinvertebrate indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| Sample Storage | | Qualify sample as suspect for all analyses |
| Holding time | | Qualify samples |
| Preservation | | Qualify samples |

#### 5.6.6.2 Analysis of Samples

Specific quality control measures are listed in Table 5.6-5 for laboratory operations. Figure 11 presents the general process for analyzing benthic invertebrate samples. Specific quality control measures are listed in Table 5.6-6 for laboratory identification operations.

**Table 5.6-5.** Laboratory Quality Control: benthic macroinvertebrate sample processing

| Check or Sample Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| SAMPLE PROCESSING (PICK AND SORT) | | | |
| Sample residuals examined by different analyst within lab | 10% of all samples completed per analyst | Efficiency of picking ≥90% | If <90%, examine all residuals of samples by that analyst and retrain analyst |

| | | | |
|---|---|---|---|
| Sorted samples sent to independent lab | 10% of all samples | Accuracy of contractor laboratory picking and identification ≥90% | If picking accuracy <90%, all samples in batch will be reanalyzed by contractor |

**Table 5.6-6:** Laboratory Quality Control: benthic macroinvertebrate taxonomic identification

| Check or Sample Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Duplicate identification by different taxonomist within lab | 10% of all samples completed per laboratory | Efficiency ≥85% | If <85%, reidentify all samples completed by that taxonomist |
| Independent identification by outside taxonomist | All uncertain taxa | Uncertain identifications to be confirmed by expert in particular taxa | Record both tentative and independent IDs |
| Use widely/commonly excepted taxonomic references | For all identifications | All keys and references used must be on bibliography prepared by another laboratory | If other references desired, obtain permission to use from Project QA Officer |
| Prepare reference collection | Each new taxon per laboratory | Complete reference collection to be maintained by each individual laboratory | Lab Manager periodically reviews data and reference collection to ensure reference collection is complete and identifications are accurate |

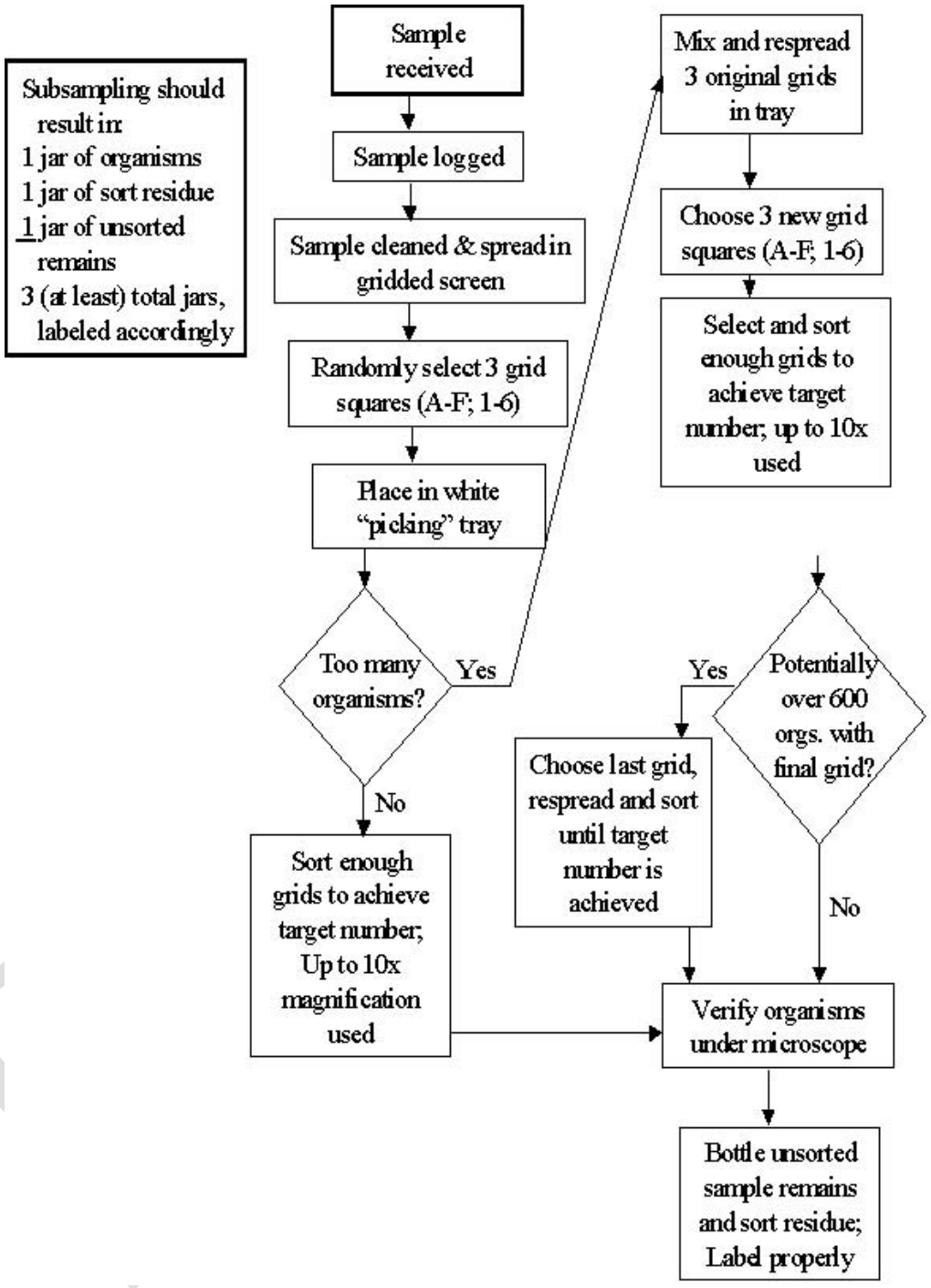


**Figure 11:** Laboratory Processing Activities for the benthic indicator

### 5.6.7 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.6-7. The Project Facilitation Team is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA project by EMAP and copied onto a CD. The CDs are transferred to the NRSA IM Coordinator (Marlys Cappaert) for entry into a centralized data base. A hard copy output of all files accompanies each data CD.

A reference specimen collection is prepared as new taxa are encountered in samples. This collection consists of preserved specimens in vials and mounted on slides and is provided to the responsible EPA laboratory as part of the analytical laboratory contract requirements. The reference collection is archived at the responsible EPA laboratory.

Sample residuals, vials, and slides are archived by each laboratory until the NRSA Project Leader has authorized, in writing, the disposition of samples. All raw data (including field data forms and bench data recording sheets) are retained in an organized fashion indefinitely or until written authorization for disposition has been received from the NRSA Project Leader.

**Table 5.6-7: Data** review, verification, and **validation quality control: benthic indicator**

| Check Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Taxonomic "reasonableness" checks | All data sheets | Genera known to occur in given stream or river conditions or geographic area | Second or third identification by expert in that taxon |

### 5.6.8 Data Analysis Plan

Specific research issues to be addressed from this year's activities and the ecological attributes or metrics associated with the benthic indicator are summarized in Table 5.6-8.

**Table 5.6-8. Research issues: benthic indicator**

| Research Issues | Design Strategy |
|---|---|
| Variance Estimates | Obtain estimates of variance components from duplicate samples and revisits to sites. |
| Indicator Development and Evaluation | Identify best set of ecological attributes or metrics that are broadly applicable to assessing biological condition and are informative as to detection and characterization of impairment. Candidate attributes are selected measures of richness, O/E, representatives of sensitive taxa. These are based on EPA's biological condition gradient attributes as part of the aquatic life use initiative. |

| | |
|---|---|
| Methods Comparability | Use standardized guidelines (from the NWQMC Methods and Data Comparability Board) for methods comparability studies (to measure precision and sensitivity along environmental and disturbance gradients), and select ecologic al attributes best suited to compare performance of methods (e.g., compositional metrics, or richness adjusted for reference). |
| Threshold Development for Assessment | Develop general expectations for each attribute (for each ecoregion) from collection of reference sites sampled with NRSA methods. Supplement with information from states and existing data where methods differences are not an issue. Combining data for an integrated assessment is based on minimizing sampling bias. Explore the use of thresholds based on % difference, e.g., 20% deviation from reference as a consistent means of evaluating biological condition across ecoregions. |
| Biological Condition | Develop an ordinal scale related to a biological condition gradient to reflect varying degrees of quality. |

## 5.7 Fish Community Structure

### 5.7.1 Introduction

Monitoring of the fish assemblage is an integral component of many water quality management programs. The assessment will measure specific attributes of the overall structure and function of the ichthyofaunal community to evaluate biological integrity and water quality.

### 5.7.2 Sampling Design

The fish sampling method is designed to provide a representative sample of the fish community, collecting all but the rarest fish inhabiting the site. It is assumed to accurately represent species richness, species guilds, relative abundance, and anomalies. The goal is to collect fish community data that will allow the calculation of an Index of Biotic Integrity (IBI) and Observed/Expected (O/E) models. Backpack or barge electrofishing is the preferred method. If electrofishing is not possible due to safety concerns, high turbidity, or extremes in conductivity, complete the "Not Fished" section of the field form and comment why.

### 5.7.3 Sampling and Analytical Methods

#### 5.7.3.1 Wadeable Streams

Streams with mean wetted widths less than 12.5 m will be electrofished in their entirety, covering all available habitats. However, the time and effort necessary to sample reaches greater than 12.5 m wide is prohibitive in the context of the survey, thus sub-sampling is required. Sub-sampling is defined by 5-10 sampling zones, each starting at a transect. In all instances electrofishing in wadeable systems should proceed in an upstream direction using a single anode. Identification and processing of fish should occur at the completion of each transect.

#### 5.7.3.2 Non-wadeable Streams

The time and effort necessary to sample the reach in its entirety is prohibitive in the context of the survey, thus sub-sampling is required. Electrofishing will occur in a downsteam direction at all habitats along alternating banks over a length of 20 times the mean channel width (5 transects - A through E). Collection of a minimum of 500 fish is required. If this target is not attained, sampling will continue until 500 individuals are captured or the downstream extent of the site (transect K) is reached. Identification and processing of fish should occur at the completion of each transect.

### 5.7.4 Quality Assurance Objectives

MQOs are given in Table 5.7-1. General requirements for comparability and representativeness are addressed in Section 2. Precision is calculated as percent efficiency, estimated from independent identifications of organisms in randomly selected samples. The MQO for

accuracy is evaluated by having individual specimens representative of selected taxa identified by recognized experts.

**Table 5.7.1.** Measurement data quality objectives: fish community indicator

| Variable or Measurement | Precision | Accuracy | Completeness |
|---|---|---|---|
| | | | |
| | | | |

NA = not applicable
[a]Taxonomic accuracy, as calculated using Equation 10 in Section 2.

### 5.7.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements.

Review all collecting permits to determine if any sampling restrictions are in effect for the site. In some cases, you may have to cease sampling if you encounter certain listed species. An experienced fisheries biologist sets up the electrofishing equipment. After selecting the initial voltage setting and pulse rate, the crew starts electrofishing. If fishing success is poor, increase the pulse width first and then the voltage to sample effectively and minimize injury and mortality. Increase the pulse rate last to minimize mortality or injury to large fish. If mortalities occur, first decrease pulse rate, then voltage, then pulse width. Fishing begins with a cleared clock to document button time. If button time is not metered, estimate it with a stop watch and flag the data.

Crews may choose to have more than one person holding a net, but no more than one person should be netting at any one time. To reduce stress and mortality, immobilized fish should be netted immediately and deposited into a live-well for processing. Process fish when fish show signs of stress (e.g., loss of righting response, gaping, gulping air, excessive mucus). Change water or stop fishing and initiate processing as soon as possible. Similarly, State- and Federally-listed threatened or endangered species or large game fish should be processed and released as they are captured. If periodic processing is required, fish should be released in a location that prevents the likelihood of their recapture. For safety, all crew members are required to wear non-breathable waders and insulated gloves. Polarized sunglasses and caps to aid vision are also required.

An experienced fisheries biologist will identify the collected fish specimens in the field. All specimens must be identified by common name as listed in Appendix D of the Field

Operations Manual. The biologist may chose to retain certain specimens for identification or verification in the laboratory. These samples are retained at the discretion of the fisheries biologist and are separate from the official voucher specimens that must be collected at 10% of each field crews' sites to be re-identified by an independent taxonomist.

Check the sample labels for all voucher and laboratory ID specimens to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the voucher sample on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the samples or if conditions occur that may affect sample integrity. Preserve all voucher samples with 10% buffered formalin and store them in a sturdy container (i.e., cooler) until shipment to the analytical laboratory. Recheck all forms for completeness and legibility. Additionally, duplicate (replicate) samples will be collected at 10% of sites sampled. A summary of Field quality control procedures for the fish community indicator is presented in Table 5.7-2.

**Table 5.7-2.** Sample collection and field processing quality control: fish community indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Set up electrofishing equipment | An experienced fisheries biologist sets up the unit. If results are poor, adjustments are made to the pulse width and voltage to sample effectively and minimize injury/mortality. | |
| Comparable effort | Reset unit clock to document button time (700 seconds per transect). If button time is not metered, estimate it with a stop watch and flag the data. | |
| Comparable effort | No more than 1 person is netting at any one time. | |
| Field Processing | Immobilized fish are netted immediately and deposited into livewell. Process before fish show signs of stress. State or federally listed threatened or endangered species or large game fish should be processed and released as they are captured. | |
| Field Processing | Fish should be released in a location that prevents the likelihood of their recapture. | |
| Field Processing | The fisheries biologist will identify specimens in the field using a standardized list of common names (App. D of the Field Operations Manual). | |

| | | |
|---|---|---|
| Sample Collection | The biologist may retain uncertain specimens for ID or verification in the laboratory. These samples are retained at the discretion of the biologist and are separate from the official voucher specimens that must be collected at 10% of each field crews' sites to be re-identified by an independent taxonomist. | |
| Sample Collection - Taxonomic QC samples | 10% of each field crews' sites are randomly selected for re-identification by an independent taxonomist. A minimum of 1 complete voucher is required for each field taxonomist and will consist of either preserved specimen(s) or digital images representative of all species in the sample, even common species. | |
| Sample Preservation | Fish retained for lab ID or vouchers are preserved with 10% buffered formalin. All personnel must read the MSDS (App D of QAPP). | |
| Safety | All crew members are required to wear insulated gloves and non-breathable waders. Caps and polarized sunglasses to aid vision are also required. | |
| Safety | Wear vinyl or nitrile gloves and safety glasses, and always work in a well-ventilated area. | |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |

#### 5.7.5.1 Sample Preservation

Fish retained for laboratory identification or as vouchers should be preserved in the field with 10% buffered formalin. The specimens should be placed in a large sample jar containing a 10% buffered formalin solution in a volume equal to or greater than the total volume of specimens. Individuals larger than 200 mm in total length should be slit along the right side of the fish in the lower abdominal cavity to allow penetration of the solution. All personnel handling 10% buffered formalin must read the MSDS (Appendix D). Formalin is a potential carcinogen and should be used with extreme caution, as vapors and solution are highly caustic and may cause severe irritation on contact with skin, eyes, or mucus membranes. Wear vinyl or nitrile gloves and safety glasses, and always work in a well-ventilated area**.**

#### 5.7.5.2 Laboratory Identification

Fish that are difficult to identify in the field are kept for laboratory identification or to verify difficult field identifications. Table 6.5-5 in the Field Operations Manual outlines the laboratory identification process and completing the Fish Collection Form. Field crews must retain the Fish Collection Form(s) for all sites until the laboratory identification

process is complete. Crews should retain the Fish verification sample – contact your regional EPA coordinator if you cannot store the samples at your facility.

#### 5.7.5.3 Voucher Specimens

Approximately 10% of each field crews' sites will be randomly pre-selected for re-identification by an independent taxonomist. A minimum of one complete voucher is required for each person performing field taxonomy and will consist of either preserved specimen(s) or digital images representative of all species in the sample, even common species. Multiple specimens per species can be used as vouchers, if necessary (i.e., to document different life or growth stages, or sexes). Note that a complete sample voucher does not mean that all individuals of each species will be vouchered, only enough so that independent verification can be achieved.

For species that are retained, specimen containers should be labeled with the sample number, site ID number, site name, and collection date. There should be no taxonomic identification labels in or on the container.

Digital images should be taken as voucher documentation for species that are recognized as Rare, Threatened, or Endangered (RTE) – they should not be harmed or killed. Very common and well-known, or very large-bodied species should also be recorded by digital images; however, these can be preserved at the discretion of the taxonomist. Labeling, within the image, should be similar to that used for preserved samples and not include taxonomic identification. Guidance for naming photo files is provided below in the photovouchering section.

#### 5.7.5.4 Photovouchering

Digital imagery should be used for fish species that cannot be retained as preserved specimens (e.g., RTE species; very large bodied; or very common). Views appropriate and necessary for an independent taxonomist to accurately identify the specimen should be the primary goal of the photography. Additional detail for these guidelines is provided in Stauffer et al. (2001), and is provided to all field crews as a handout.

The recommended specifications for digital images to be used for photovouchering include: 16-bit color at a minimum resolution of 1024x768 pixels; macro lens capability allowing for images to be recorded at a distance of less than 4 cm; and built-in or external flash for use in low-light conditions. Specimens should occupy as much of the field of view as possible, and the use of a fish board is recommended to provide a reference to scale (i.e., ruler or some calibrated device) and an adequate background color for photographs. Information on Station ID, Site Name, Date and a unique species ID (i.e., A, B, C, etc.) should also be captured in the photograph, so that photos can be identified if file names become corrupted. All photovouchered species should have at least a full-body photo (preferably of the left side of the fish) and other zoom images as necessary for individual species, such as lateral line, ocular/oral orientation, fin rays, gill arches, or others. It may also be necessary to photograph males, females, or juveniles.

Images should be saved in medium- to high-quality jpeg format, with the resulting file name of each picture noted one the Fish Collection Form. It is important that time and date stamps are accurate as this information can also be useful in tracking the origin of photographs. It is recommended that images stored in the camera be transferred to a PC or storage device at the first available opportunity. At this time the original file should be renamed to follow the logic presented below:

**F01_CT003_20080326.jpg**

where F=fish, 01=tag number, CT003=state (Connecticut) and site number, and 20080326=date (yyyymmdd).

Field crews should maintain files for the duration of the sampling season. Notification regarding the transfer of all images to the existing database will be provided at the conclusion of the sampling.

### 5.7.6 Quality Control Procedures: Laboratory Operations (Voucher Specimens)

#### 5.7.6.1 Sample Receipt and Processing

QC activities associated with sample receipt and processing are presented in Table 5.7-3. The communications center and information management staff are notified of sample receipt and any associated problems as soon as possible after samples are received.

**Table 5.7-3.** Sample receipt and processing quality control: fish community indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Sample Log-in | Upon receipt of a sample shipment, laboratory personnel check the condition and identification of each sample against the sample tracking record. | Discrepancies, damaged, or missing samples are reported to the IM staff and indicator lead |
| Sample Storage | | Qualify sample as suspect for all analyses |
| Holding time | | Qualify samples |
| Preservation | | Qualify samples |

#### 5.7.6.2 Analysis of Samples

Specific quality control measures are listed in Table 5.7-4 for laboratory operations.

**Table 5.7-4:** Laboratory Quality Control: fish voucher taxonomic identification

| Check or Sample Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Independent identification by outside taxonomist | Complete voucher colection for 10% of all sites | Uncertain identifications to be confirmed by expert in particular taxa | If <85%, reidentify all samples completed by that taxonomist |
| Use widely/commonly excepted taxonomic references | For all identifications | All keys and references used must be on bibliography prepared by another laboratory | If other references desired, obtain permission to use from Project QA Officer |

## 5.8 Physical Habitat Quality

### 5.8.1 Introduction

Naturally occurring differences in physical habitat structure and associated hydraulic characteristics among surface waters contributes to much of the observed variation in species composition and abundance within a zoogeographic province. Structural complexity of aquatic habitats provides the variety of physical and chemical conditions to support diverse biotic assemblages and maintain long-term stability. Anthropogenic alterations of riparian physical habitat, such as channel alterations, wetland drainage, grazing, agricultural practices, weed control, and streambank modifications such as revetments or development, generally act to reduce the complexity of aquatic habitat and result in a loss of species and ecosystem degradation.

For the NRSA, indicators derived from data collected on physical habitat quality will be used to help explain or characterize stream and river conditions relative to biological response and trophic state indicators. Specific groups of physical habitat attributes important in stream and river ecology include: channel dimensions, gradient, substrate; habitat complexity and cover; riparian vegetation cover and structure; anthropogenic alterations; and channel-riparian interaction (Kaufmann, 1993). Overall objectives for this indicator are to develop quantitative and reproducible indices, using both multivariate and multimetric approaches, to classify streams and rivers and to monitor biologically relevant changes in habitat quality and intensity of disturbance.

### 5.8.2 Sampling Design

As the physical habitat indicator is based on field measurements and observations, there is no sample collection associated with this indicator. Field crews are provided with 1:24,000 maps with the midpoint (index site) of the stream reach marked. At NRSA sites, eleven cross-sectional measurement transects are spaced at equal intervals proportional to baseflow channel width, thereby scaling the sampling reach length and resolution in proportion to stream and river size. A systematic spatial sampling design is used to minimize bias in the selection of the measurement sites. Additional measurements are made at equally spaced intervals between the cross-sectional sites.

### 5.8.3 Sampling Methodologies

*Field Measurements:* Field measurements, observations, and associated methodology for the protocol are summarized in Table 5.8-1. Detailed procedures for completing the protocols are provided in the field operations manual; equipment and supplies required are also listed. All measurements and observations are recorded on standardized forms which are later entered in to the central EMAP surface waters information management system at WED-Corvallis.

There are no sample collection or laboratory analyses associated with the physical habitat measurements.

**Table 5.8-1.** Field measurement methods: physical habitat indicator

| Variable or Measurement | Units | QA | Summary of Method | References |
|---|---|---|---|---|
| THALWEG PROFILE | | | | |
| Thalweg depth | cm | C | Measure max depth at 100-150 points for wadeable or 200 points for non-wadeable along reach with surveyor's rod or sonar equipment | |
| Wetted width | 0.1m | C | Measure wetted width with range finder or measuring tape on perpendicular line to mid-channel line | |
| Habitat class | none | N | Visually estimate channel habitat using defined class descriptions | Frissell et al, 1986 |
| WOODY DEBRIS TALLY | | | | |
| Large woody debris | # of pieces | N | Use pole drag and visually estimate amount of woody debris in baseflow channel using defined class descriptions | Robison and Beschta, 1990 |
| CHANNEL AND RIPARIAN CROSS-SECTIONS | | | | |
| Slope and bearing | %/ degrees | C | Backsight between cross-section stations using clinometer, rangefinder compass, & tripod | Robison & Kaufman n, in prep.; Stack, 1989 |
| Substrate size | mm | C | At 5 points on cross section, estimate size of one selected particle using defined class descriptions | Wollman, 1954; Bain et al, 1985; Plafkin et al, 1989 |
| Bank angle | degrees | N | Use clinometer and surveyors rod to measure angle | Platts et al, 1983 |
| Bank incision | 0.1m | N | Visually estimate height from water surface to first terrace of floodplain | |
| Bank undercut | cm | N | Measure horizontal distance of undercut | |
| Bankful width | 0.1m | N | Measure width at top of bankful height | |
| Bankful height | 0.1m | N | Measure height from water surface to estimated water surface during bankful flow | |
| Canopy cover | points of | C | Count points of intersection on densiometer at specific points and directions on cross-section | Lemmon, 1957; Mulvey et al, |

| | | | | |
|---|---|---|---|---|
| | intersection | | | 1992 |
| Riparian vegetation structure | percent | N | Observations of ground cover, understory, and canopy types and coverage of area 5 m on either side of cross section and 10 m back from bank | |
| Fish cover, algae, macrophytes | percent | C | Visually estimate in-channel features 5 m on either side of cross section | |
| Human influence | none | C | Estimate presence/absence of defined types of anthropogenic features | |
| STREAM DISCHARGE | | | | |
| Discharge | m/s or L/min. | N | Velocity-Area method, Portable Weir method, timed bucket discharge method | Linsley et al, 1982 |

### 5.8.4 Quality Assurance Objectives

Measurement data quality objectives (measurement DQOs or MQOs) are given in Table 5.8-2. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.8-2 represent the maximum allowable criteria for statistical control purposes. Precision is determined from results of revisits by a different crew (field measurements) and by duplicate measurements by the same crew on a different day.

The completeness objectives are established for each measurement *per site type* (e.g., NRSA sites, revisit sites, state comparability sites). Failure to achieve the minimum requirements for a particular site type results in regional population estimates having wider confidence intervals. Failure to achieve requirements for repeat and annual revisit samples reduces the precision of estimates of index period and annual variance components, and may impact the representativeness of these estimates because of possible bias in the set of measurements obtained.

**Table 5.8-2.** Measurement data quality objectives: physical habitat indicator

| Variable or Measurement | Precision | Accuracy | Completeness |
|---|---|---|---|
| Field Measurements and Observations | ±10%* | NA | 90% |
| Map-Based Measurements | ±10% | NA | 100% |

NA = not applicable *Not for RBP measures

### 5.8.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements. Specific quality control measures are listed in Table 5.8-3 for field measurements and observations.

**Table 5.8-3. Field quality control: physical habitat indicator**

| Check Description | Frequency | Acceptance Criteria | Corrective Actions |
|---|---|---|---|
| Check totals for cover class categories (vegetation type, fish cover) | Each transect | Sum must be reasonable (best professional judgement) | Repeat observations |
| Check completeness of thalweg depth | Each site | Depth measurements for all sampling points | Obtain best estimate of depth where |

| | | | |
|---|---|---|---|
| measurements | | | actual measurement not possible |
| Check calibration of multiprobe | Prior to each sampling day | Specific to instrument | Adjust and recalibrate, redeploy gear |

### 5.8.6 Quality Control Procedures: Laboratory Operations

There are no laboratory operations associated with this indicator.

### 5.8.7 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.8-4. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. All raw data (including all standardized forms and logbooks) are retained in an organized fashion for seven years or until written authorization for disposition has been received from the NRSA Project Coordinator.

**Table 5.8-4.** Data validation quality control: physical habitat indicator

| Check Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Estimate precision of measurements based on repeat visits by different crews | At least 2 teams visit stream and river 1 time each at 10% of streams and rivers (may be same team or different teams) | Measurements should be within 10 percent | Review data for reasonableness; Determine if acceptance criteria need to be modified |

## 5.9 Fish Tissue

### 5.9.1 Introduction

Fish are time-integrating indicators of persistent pollutants, and contaminant bioaccumulation in fish tissue has important human and ecological health implications. Contaminants in fish pose risks to human consumers and to piscivorous wildlife. The NRSA fish tissue indicator will provide information on the national distribution of selected persistent, bioaccumulative, and toxic (PBT) chemical residues (e.g., mercury and organochlorine pesticides) in predator fish species from large (non-wadeable) streams and rivers of the conterminous United States. Recent studies show that an emerging group of contaminants – pharmaceuticals and personal care products (PPCPs) – can persist through the wastewater treatment process and occur in municipal effluent, surface water, and sediments. However, data on the accumulation of PPCPs in fish are scarce. NRSA fish tissue samples will be used to address this data gap. Samples collected from a national statistical subset of NRSA urban sites (approximately 150 sites) located on large (non-wadeable) rivers will be analyzed for PPCPs.

The fish tissue indicator procedures are based on EPA's *National Study of Chemical Residues in Lake Fish Tissue* (USEPA 2000a) and EPA's *Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1 (Third Edition)* (USEPA 2000b).

### 5.9.2 Sampling Design

The NRSA crews will collect fish for the tissue indicator from all non-wadeable study reaches sampled for the fish community structure indicator (Section 5.8). Fish tissue samples must consist of a composite of fish (i.e., five individuals of one predator species that will collectively provide greater than 500 grams of fillet tissue) from each site. Tissue sampling may require additional effort (temporally and/or spatially) beyond that of the fish community structure sampling. Fish retained for the tissue indicator may be collected from anywhere between site transects A and K.

Field teams will consist of one experienced fisheries biologist and one field technician. The experienced on-site fisheries biologist will select the most appropriate electrofishing gear type(s) for a particular site. The appropriate sampling equipment will be based on the size/depth of each site, and deployment will target recommended predator species (Table 5.9.1). Accurate taxonomic identification is essential to prevent mixing of species within composites. Five fish will be collected per composite at each site, all of which must be large enough to provide sufficient tissue for analysis (i.e., 500 grams of fillets, collectively). Fish in each composite must all be of the same species, satisfy legal requirements of harvestable size (or be of consumable size if there are no harvest limits), and be of similar size so that the smallest individual in the composite is no less that 75% of the total length of the largest individual. If the recommended target species are unavailable, the on-site fisheries biologist will select an alternative species (i.e., a predator species that is commonly consumed in the study area, with specimens of harvestable or consumable size, and in sufficient numbers to yield a composite).

**Table 5.9.1.** Recommended Target Species for Fish Tissue Collection (In Order of Preference) at non-wadeable sites

| Predator/Gamefish Species (in order of preference) | Family name | Common name | Scientific name | Length Guideline (Estimated Minimum) |
|---|---|---|---|---|
| | | Largemouth bass | *Micropterus salmoides* | ~280 mm |
| | | Smallmouth bass | *Micropterus dolomieu* | ~300 mm |
| | | Black crappie | *Pomoxis nigromaculatus* | ~330 mm |
| | *Centrarchidae* | White crappie | *Pomoxis annularis* | ~330 mm |
| | | Walleye/sauger | *Sander vitreus /S. canadensis* | ~380 mm |
| | *Percidae* | Yellow perch | *Perca flavescens* | ~330 mm |
| | *Percichthyidae* | White bass | *Morone chrysops* | ~330 mm |
| | *Esocidae* | Northern pike | *Esox lucius* | ~430 mm |
| | | Lake trout | *Salvelinus namaycush* | ~400 mm |
| | | Brown trout | *Salmo trutta* | ~300 mm |
| | | Rainbow trout | *Oncorhynchus mykiss* | ~300 mm |
| | *Salmonidae* | Brook trout | *Salvelinus fontinalis* | ~330 mm |

### 5.9.3 Sampling and Analytical Methodologies

The fish tissue sample collection schedule will be consistent with the requirements specified in this QAPP for all other NRSA indicators with the following exception: replicate fish tissue samples will be collected at revisit sites only during the first round of sampling. The sampling teams are responsible for providing fisheries sampling gear and sampling vessels. Fish selected for compositing should be rinsed in ambient water, handled using clean nitrile gloves, and placed in clean holding containers (e.g., livewells or buckets). Each fish of the selected target species should be measured to determine total body length (i.e., length from the anterior-most part of the fish to the tip of the longest caudal fin ray when the lobes of the caudal fin are depressed dorsoventrally) recorded in millimeters. When sufficient numbers of the target species have been identified to make up a suitable composite (i.e., five individuals meeting the criteria presented above), the species name, specimen lengths, and all other site sampling information should be recorded on the fish tissue field form.

After initial processing to determine species and size, each of the five fish found to be suitable for the composite sample will be individually wrapped in extra heavy-duty aluminum foil (provided by EPA as solvent-rinsed, oven-baked sheets). A sample identification label will be completed for each fish specimen. Each foil-wrapped fish and sample identification label will be placed into waterproof plastic tubing that will be cut to fit the specimen (i.e., heavy duty food grade polyethylene tubing provided by EPA), and each end of the tubing will be sealed with a plastic cable tie. All five individually-wrapped specimens from each site will be placed in a large plastic composite bag and sealed with another cable tie.

EPA will provide fish tissue sample packing and shipping supplies (with the exception of dry ice). A list of equipment and expendable supplies is provided in the NRSA Field Operations Manual. Following collection, wrapping, and labeling, samples should be immediately placed on dry ice for shipment. If samples will be carried back to an interim location to be frozen before shipment, wet ice can be used to transport the samples in coolers to that location. Each sampling team will ship all fish tissue samples in coolers on dry ice (i.e., a recommended 50 pounds per cooler) via priority overnight delivery service to a sample control center designated by EPA. All cooler vent holes must be taped open to allow gasses to escape, and the cooler lids will be sealed with a custody seal that has been signed and dated by the collector. The time of sample collection, relinquishment by the sample team, and time of their arrival at the sample preparation laboratory must be recorded on the NRSA chain-of-custody form.

### 5.9.4 Quality Assurance Objectives

The relevant quality objectives for fish tissue sample collection activities are primarily related to sample handling issues. Types of field sampling data needed for the fish tissue indicator are listed in Table 5.9.2. Methods and procedures described in this QAPP and the NRSA Field Operations Manual are intended to reduce the magnitude of the sources of uncertainty (and their frequency of occurrence) by applying:

- standardized sample collection and handling procedures, and
- use of trained scientists to perform the sample collection and handling activities.

**Table 5.9.2.** Field Data Types: Fish Tissue Indicator

| Variable or Measurement | Measurement Endpoint or Unit |
|---|---|
| Fish specimen | Species-level taxonomic identification |
| Fish length | Millimeters (mm), total length |
| Composite classification | Composite identification number |
| Specimen count classification | Specimen number |

### 5.9.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All fish tissue sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements. Specific quality control measures are listed in Table 5.9-3 for field measurements and observations.

**Table 5.9-3. Field quality control: fish tissue indicator**

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Set up electrofishing equipment | An experienced fisheries biologist sets up the unit. If results are poor, adjustments are made to the pulse width and voltage to sample effectively and minimize injury/mortality. | |
| Field Processing | The fisheries biologist will identify specimens in the field using a standardized list of common names (App. D of the Field Operations Manual). | |
| Sample Collection | The biologist will retain 5 specimens of the same species to form the composite sample. | |
| Sample Collection | The length of the smallest fish must be at least 75% of the length of the longest fish. | |

### 5.9.7 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.9-4. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. All raw data (including all standardized forms and logbooks) are retained in an organized fashion for seven years or until written authorization for disposition has been received from the NRSA Project Coordinator. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA project by EMAP and copied onto a CD. The CDs are transferred to the NRSA IM Coordinator (Marlys Cappaert) for entry into a centralized data base. A hard copy output of all files accompanies each data CD.

**Table 5.9-4.** Data validation quality control: fish tissue indicator

| Check Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| Duplicate sampling | Duplicate composite samples collected at 10% of sites | Measurements should be within 10 percent | Review data for reasonableness; determine if acceptance criteria need to be modified |
| Taxonomic "reasonableness" checks | All data sheets | Genera known to occur in stream or river conditions or geographic area | Second or third identification by expert in that taxon |
| Composite validity check | All composites | Each composite sample must have 5 fish | Indicator lead will review composite data and |

| | | | |
|---|---|---|---|
| | | of the same species | advise the lab before processing begins |
| 75% rule | All composites | Length of smallest fish in the composite must be at least 75% of the length of the longest fish. | Indicator lead will review composite data and advise the lab before processing begins |

### 5.9.8 Data Analysis Plan

Fish tissue concentration data from laboratory analysis of the fish composite samples will be reported as percentiles, including the 50$^{th}$ percentile or median concentration, for each target chemical. Cumulative distribution of fish tissue concentrations for the sampled population of sites will be estimated using a procedure described by Diaz-Ramos et al. (1996) entitled, "Estimation Method 1: Cumulative Distribution Function for Proportion of a Discrete or an Extensive Resource." The estimated proportion ($p_c$) below a specific value for a concentration (C) is:

$$p_C = \frac{\sum_{i=1}^{n} w_i * x_i}{\sum_{i=1}^{n} w_i}$$

where: $x_i$ = 1 if concentration for $i^{th}$ lake is below C and equals 0 otherwise,
$w_i$ = the adjusted weight for $i^{th}$ lake, and
n = total number of lakes sampled.

A cumulative distribution function (CDF) offers an approach to displaying statistical data that correlates the results to the sampled population. In technical terms, a CDF characterizes the probability distribution of a random variable. For the tissue indicator, the random variable is the concentration of a particular chemical in fish tissue.

Variance estimates will be derived using the local neighborhood variance estimator described by Stevens and Olsen (2003 and 2004). To complete these analyses, R statistical software (R Development Core Team 2004) and an R contributed library will be utilized for probability survey population estimation (spsurvey). The R library is available online at the following Internet address: http://www.epa.gov/nheerl/arm/analysispages/software.

## 5.10 Fecal Indicator: Enterococci

### 5.10.1 Introduction

The primary function of collecting water samples for Pathogen Indicator Testing is to provide a relative comparison of fecal pollution indicators for national rivers and streams. The concentration of *Enterococci* (the current bacterial indicator for fresh and marine waters) in a water body correlates with the level of more infectious gastrointestinal pathogens present in the water body. While some Enterococci are opportunistic pathogens among immuno-compromised human individuals, the presence of Enterococci is more importantly an indicator of the presence of more pathogenic microbes (bacteria, viruses and protozoa) associated with human or animal fecal waste. These pathogens can cause waterborne illness in bathers and other recreational users through exposure or accidental ingestion. Disease outbreaks can occur in and around beaches that become contaminated with high levels of pathogens. Therefore, measuring the concentration of pathogens present in river and stream water can help assess comparative human health concerns regarding recreational use.

In this survey, a novel, Draft EPA Quantitative PCR Method (1606) will be used to measure the concentration of genomic DNA from the fecal indicator group *Enterococcus* in the water samples. While neither federal or state Water Quality Criteria (standards) have been formally established for the level of *Enterococcus* DNA in a sample, epidemiological studies (Wade *et al.* 2005) have established a strong correlation between *Enterococcus* DNA levels and the incidence of high-credible gastrointestinal illness (HCGI) among swimmers. The *Enterococcus* qPCR results will serve as an estimate of the concentration of total (culturable and non-culturable) Enterococci present in the surveyed rivers and streams for the purpose of comparative assessment. This study also has the potential to yield invaluable information about the inhibitory effects of water matrices from the different regions of the nation upon the qPCR assay.

### 5.10.2 Sampling Design

A single “pathogen” water sample will be collected from one sampling location approximately 1 m offshore, in conjunction with the final physical habitat sampling station location.

### 5.10.3 Sampling Methods

*Sample Collection*: At the final physical habitat shoreline station (located approximately 1 m off shore), a single 1-L water grab sample is collected approximately 6-12 inches below the surface of the water. Detailed procedures for sample collection and handling are described in the Field Operations Manual. Pathogen samples must be filtered and the filters must be folded and frozen in vials within 6 hours of collection.

*Analysis:* Pathogen samples are filter concentrated, then shipped on dry ice to the New England Regional Laboratory where the filter retentates are processed, and the DNA extracts are analyzed using Quantitative Polymerase Chain Reaction (qPCR), a genetic method that quantifies a DNA target via a fluorescently tagged probe, based on methods developed by the USEPA National Exposure Research Laboratory. Detailed procedures

are contained in the laboratory operations manual. Table 5.10-1 summarizes field and analytical methods for the pathogen indicator.

**Table 5.10-1.** Field and laboratory methods: pathogen indicator (Enterococci)

| Variable or Measurement | QA Class | Expected Range and/or Units | Summary of Method | References |
|---|---|---|---|---|
| Sample Collection | C | NA | Sterile sample bottle submerged to collect 250-mL sample 6-12" below surface at 10m from shore | NRSA Field Operations Manual 2008 |
| Sub-sampling | N | NA | 2 x 50-mL sub-samples poured in sterile 50-mL tube after mixing by inversion 25 times. | NRSA Laboratory Methods Manual 2008 |
| Sub-sample (& Buffer Blank) Filtration | N | NA | Up to 50-mL sub-sample filtered through sterile polycarbonate filter. Funnel rinsed with minimal amount of buffer. Filter folded, inserted in tube then frozen. | NRSA Lab Methods Manual 2008 |
| Preservation & Shipment | C | -40C to +40 C | Batches of sample tubes shipped on dry ice to lab for analysis. | NRSA Lab Methods Manual 2008 |
| DNA Extraction (Recovery) | C | 10-141% | Bead-beating of filter in buffer containing Extraction Control (SPC) DNA. DNA recovery measured | EPA Draft Method 1606 *Enterococcus* qPCR |
| Method 1606 (*Enterococcus* & SPC qPCR) | C | <60 (RL) to >100,000 ENT CCEs/100-mL | 5-uL aliquots of sample extract are analyzed by ENT & Sketa qPCR assays along with blanks, calibrator samples & standards. Field and lab duplicates are analyzed at 10% frequency. Field blanks analyzed at end of testing only if significant detections observed. | EPA Draft Method 1606 *Enterococcus* qPCR<br><br>NERL NLPS2007 qPCR Analytical SOP |

C = critical, N = non-critical quality assurance classification.

### 5.10.4 Quality Assurance Objectives

Measurement quality objectives (MQO) are given in table 5.10-2. General requirements for comparability and representativeness are addressed in Section 2. Precision is calculated as percent efficiency, estimated from independent identifications of organisms in randomly selected samples. The MQO for accuracy is evaluated by having individual specimens representative of selected taxa identified by recognized experts.

**Table 5.10-2.** Measurement data quality objectives: Pathogen-Indicator DNA Sequences

| Variable or Measurement* | Method Precision | Method Accuracy | Completeness |
|---|---|---|---|
| SPC & ENT DNA sequence numbers of Calibrators & Standards by AQM | RSD=50% | 50% | 95% |
| ENT CCEs by dC*t* RQM | RSD = 70% | 35% | 95% |
| ENT CCEs by ddC*t* RQM | RSD = 70% | 50% | 95% |

*AQM = Absolute Quantitation Method; RQM = Relative Quantitation Method;
SPC = Sample Processing Control (Salmon DNA / Sketa); CCEs = Calibrator Cell Equivalents

### 5.10.5 Quality Control Procedures: Field Operations

Field data quality is addressed, in part, by application and consistent performance of valid procedures documented in the standard operating procedures detailed in the NRSA Field Operations Manual. That quality is enhanced by the training and experience of project staff and documentation of sampling activities. This QAPP, the NRSA Field Operations Manual, and training materials will be distributed to all field sampling personnel. Training sessions will be conducted by EPA to distribute and discuss project materials. All fish tissue sampling teams will be required to view the training materials, read the QAPP, and verify that they understand the procedures and requirements. Specific quality control measures are listed in Table 5.10-3 for field measurements and observations.

It is important that the sample container be completely sterilized and remain unopened until samples are ready to be collected. Once the sample bottles are lowered to the desired depth (6-12 in. below the surface), the sample bottles may then be opened and filled. After filling the 1-L bottle check the label to ensure that all written information is complete and legible. Place a strip of clear packing tape over the label and bar code, covering the label completely. Record the bar code assigned to the pathogen sample on the Sample Collection Form. Enter a flag code and provide comments on the Sample Collection Form if there are any problems in collecting the sample or if conditions occur that may affect sample integrity. All samples should be placed in coolers and maintained on ice during transport to the laboratory and maintained at 1–4°C during the time interval before they are filtered for analysis. Recheck all forms and labels for completeness and legibility.

Field blanks and duplicates will be collected at 10% of sites sampled. In addition, each field crew should collect a blank sample over the course of the survey as a check on each crew's aseptic technique and the sterility of test reagents and supplies.

**Table 5.10-3.** Sample collection and field processing quality control: fecal indicator

| Quality Control Activity | Description and Requirements | Corrective Action |
|---|---|---|
| Check integrity of sample containers and labels | Clean, intact containers and labels | Obtain replacement supplies |
| Sterility of sample containers | Sample collection bottle and filtering apparatus are sterile and must be unopened prior to sampling. Nitrile gloves must be worn during sampling and filtering | |
| Sample Collection | Collect sample at the last transect to minimize holding time before filtering and freezing | |
| Sample holding | Sample is held in a cooler on wet ice until filtering | |
| Field Processing | Sample is filtered and filters are frozen on dry ice within 6 hours of collection | |
| Duplicate samples | Duplicate samples must be collected at 10% of sites | |
| Field Blanks | Field blanks must be filtered at 10% of sites | |

### 5.10.6 Quality Control Procedures: Laboratory Operations

Specific quality control measures are listed in Table 5.10-4 for laboratory operations.

**Table 5.10-4.** Laboratory Quality Control: Pathogen-Indicator DNA Sequences

| Check or Sample Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|
| SAMPLE PROCESSING | | | |
| Re-process sub-samples (Lab Duplicates) | 10% of all samples completed per laboratory | Percent Congruence <70% RSD | If >70%, re-process additional sub-samples |
| qPCR ANALYSIS | | | |

| | | | |
|---|---|---|---|
| Duplicate analysis by different biologist within lab | 10% of all samples completed per laboratory | Percent Congruence ≤70% RSD | If >70%, determine reason and if cause is systemic, re-analyze all samples in question. |
| Independent analysis by external laboratory | None | Independent analysis TBD | Determine if independent analysis can be funded and conducted. |
| Use single stock of *E. faecalis* calibrator | For all qPCR calibrator samples for quantitation | All calibrator sample C*p* (C*t*) must have an RSD ≤ 50%. | If calibrator C*p* (C*t*) values exceed an RSD value of 50% a batch's calibrator samples shall be re-analyzed and replaced with new calibrators to be processed and analyzed if RSD not back within range. |
| DATA PROCESSING & REVIEW | | | |
| 100% verification and review of qPCR data | All qPCR amplification traces, raw and processed data sheets | All final data will be checked against raw data, exported data, and calculated data printouts before entry into LIMS and upload to Corvallis, OR database. | Second tier review by contractor and third tier review by EPA. |

### 5.10.7 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.10-5. The Indicator Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members. All raw data (including all standardized forms and logbooks) are retained in an organized fashion for seven years or until written authorization for disposition has been received from the NRSA Project Coordinator. Once data have passed all acceptance requirements, computerized data files are prepared in a format specified for the NRSA project by EMAP and copied onto a CD. The CDs are transferred to the NRSA IM Coordinator (Marlys Cappaert) for entry into a centralized data base. A hard copy output of all files accompanies each data CD.

**Table 5.10-5.** Data validation quality control: fecal indicator

| Check Description | Frequency | Acceptance Criteria | Corrective Action |
|---|---|---|---|

| | | | |
|---|---|---|---|
| Duplicate sampling | Duplicate composite samples collected at 10% of sites | Measurements should be within 10 percent | Review data for reasonableness; determine if acceptance criteria need to be modified |
| Field filter blanks | Field blanks filtered at 10% of sites | Measurements should be within 10 percent | Review data for reasonableness; determine if acceptance criteria need to be modified |
| | | | |
| | | | |

# 6.0 FIELD AND BIOLOGICAL LABORATORY QUALITY EVALUATION AND ASSISTANCE VISITS

No national program of accreditation for biological sample collections and processing currently exists. However, national standards of performance and audit guidance for biological laboratories are being considered by the National Environmental Laboratory Accreditation Conference (NELAC). For this reason, a rigorous program of field and laboratory evaluation and assistance visits has been developed to support the National Rivers and Streams Assessment Program.

Procedural review and assistance personnel are trained to the specific implementation and data collection methods detailed in the NRSA field operations manual. Plans and checklists for field evaluation and assistance visit have been developed to reinforce the specific techniques and procedures for both field and laboratory applications. The plans and checklists are included in this section and describe the specific evaluation and corrective action procedures.

It is anticipated that evaluation and assistance visits will be conducted with each Field Team early in the sampling and data collection process, and that corrective actions will be conducted in real time. These visits provide a basis for the uniform evaluation of the data collection techniques, and an opportunity to conduct procedural reviews as required to minimize data loss due to improper technique or interpretation of program guidance. Through uniform training of field crews and review cycles conducted early in the data collection process, sampling variability associated with specific implementation or interpretation of the protocols will be significantly reduced. The field evaluations, while performed by a number of different supporting collaborator agencies and participants, will be based on the uniform training, plans, and checklists. This review and assistance task will be conducted for each unique crew collecting and contributing data under this program; hence no data will be recorded to the project database that were produced by an 'unaudited' process, or individual.

Similarly, laboratory evaluation and assistance visits will be conducted early in the project schedule and soon after sample processing begins at each laboratory to ensure that specific laboratory techniques are implemented consistently across the multiple laboratories generating data for the program. Laboratory evaluation plans and checklists have been developed to ensure uniform interpretation and guidance in the procedural reviews. These laboratory visits are designed such that full corrective action plans and remedies can be implemented in the case of unacceptable deviations from the documented procedures observed in the review process without recollection of samples.

The Field and Laboratory Evaluation and Assistance Visit Plans are described in sections 6.1 and 6.2.

## 6.1 National Rivers and Streams Assessment Field Quality Evaluation and Assistance Visit Plan

**Evaluators: One or more designated EPA or Contractor staff members who are qualified (i.e., have completed training) in the procedures of the NRSA field**

**sampling operations.**

**To Evaluate: Field Sampling Teams during sampling operations on site.**
**Purpose: To identify and correct deficiencies during field sampling operations.**

1. Tetra Tech and GLEC project staff will review the Field Evaluation and Assistance Visit Plan and Check List with each Evaluator during field operations training sessions.
2. The Tetra Tech and GLEC QA Officer or authorized designee will send a copy of the final Plan and the final Check List pages, envelopes to return the Check Lists, a clipboard, pens, and the NRSA *Quality Assurance Project Plan* and *Field Operations Manua*l to each participating Evaluator.
3. Each Evaluator is responsible for providing their own field gear sufficient to accompany the Field Sampling Teams (e.g., protective clothing, sunscreen, insect repellent, hat, hip boots or waders, water bottle, food, back pack, cell phone) during a complete sampling cycle. Schedule of the Field visits will be made by the Evaluator in consultation with the Tetra Tech or GLEC QA Officer and respective Field Crew Leader. **Evaluators should be prepared to spend additional time in the field if needed (see below).**
4. Tetra Tech, GLEC, and the Regional Monitoring Coordinators will arrange the schedule of visitation with each Field Team, and notify the Evaluators concerning site locations, where and when to meet the team, and how to get there. Ideally, each Field Team will be evaluated within the first two weeks of beginning sampling operations, so that procedures can be corrected or additional training provided, if needed. EPA Evaluators will visit Tetra Tech and GLEC Field Teams. Any EPA or Contractor Evaluator may visit State Field Teams.
5. A Field Team for the NRSA consists of a four-person crew where, at a minimum, the Field Crew Leader and one additional crew member is fully trained.
6. If members of a Field Team change, and a majority (i.e., two) of the members have not been evaluated previously, the Field Team must be evaluated again during sampling operations as soon as possible to ensure that all members of the Field Team understand and can perform the procedures.
7. The Evaluator will view the performance of a team through one complete set of sampling activities as detailed on the Field Evaluation and Assistance Check List.

a. Scheduling might necessitate starting the evaluation midway on the list of tasks at a site, instead of at the beginning. In that case, the Evaluator will follow the team to the next site to complete the evaluation of the first activities on the list.
b. If the Team misses or incorrectly performs a procedure, the Evaluator will note this on the checklist and *immediately point this out so the mistake can be corrected on the spot*. The role of the Evaluator is to provide additional training and guidance so that the procedures are being performed consistent with the *Field Operations Manual*, all data are recorded correctly, and paperwork is properly completed at the site.
c. When the sampling operation has been completed, the Evaluator will review the results of the evaluation with the Field Team before leaving the site (if practicable), noting positive practices and problems, weaknesses [might affect data quality], and deficiencies [would adversely affect data quality]). The Evaluator will ensure that the Team understands the findings and will be able to perform the procedures properly in the future.

d. The Evaluator will record responses or concerns, if any, on the Field Evaluation and Assistance Check List.

e. If the Evaluator's findings indicate that the Field Team is not performing the procedures correctly, safely, or thoroughly, the Evaluator must continue working with this Field Team until certain of the Team's ability to conduct the sampling properly so that data quality is not adversely affected.

f. If the Evaluator finds major deficiencies in the Field Team operations (e.g., less than three members, equipment or performance problems) the Evaluator must contact one of the following QA officials:

Dr. Esther Peters, Tetra Tech QA Officer (703-385-6000)
Ms. Robin Silva-Wilkinson, GLEC QA Officer (231-941-2230)
Mr. Richard Mitchell, EPA NRSA Project QA Officer (202-566-0644)

The QA official will contact the Project Implementation Coordinator ( Ellen Tarquinio – 202-566-2267 ) to determine the appropriate course of action.

Data records from sampling sites previously visited by this Field Team will be checked to determine whether any sampling sites must be redone.

g. Complete the Field Evaluation and Assistance Check List, including a brief summary of findings, and ensure that all Team members have read this and signed off before leaving the Team.

8. The Evaluator will electronically scan and make a photocopy of the Field Evaluation and Assistance Check List. The Evaluator will retain the photocopied checklist, and email the scanned file and send the original checklist to

Richard Mitchell
USEPA Office of Wetlands, Oceans, and Watersheds
1200 Pennsylvania Avenue (4503-T)
Washington, DC 20460-0001
(202)-566-0644

### 6.2 National Rivers and Streams Assessment Laboratory Quality Evaluation and Assistance Visit Plan

**Evaluators: One or more designated Contractor staff members who are qualified (i.e., have completed training) in the procedures of the NRSA biological laboratory operations.**
**To Evaluate: Biological laboratories performing subsampling, sorting, and taxonomic procedures to analyze collected stream and river samples.**
**Purpose: To identify and correct deficiencies during laboratory operations.**

1. Tetra Tech project staff will review the Laboratory Evaluation and Assistance Visit Plan and Check List with each Evaluator prior to conducting laboratory evaluations.

2. The Tetra Tech QA Officer or authorized designee will send a copy of the final Plan and final Check List pages, envelopes to return the Check Lists, a clipboard, pens, and the NRSA *Quality Assurance Project Plan* and *Laboratory Method* Manual to each participating Evaluator.
3. Schedule of lab visits will be made by the Evaluator in consultation with the Tetra Tech QA Officer and the respective Laboratory Supervisor Staff. **Evaluators should be prepared to spend additional time in the laboratory if needed (see below)**.
4. Tetra Tech, GLEC, and the Regional Monitoring Coordinators will arrange the schedule of visitation with each participating Laboratory, and notify the Evaluators concerning site locations, where and when to visit the laboratory, and how to get there. Ideally, each Laboratory will be evaluated within the first two weeks following initial receipt of samples, so that procedures can be corrected or additional training provided, if needed.
5. The Evaluator will view the performance of the laboratory sorting process and QC Officer through one complete set of sample processing activities as detailed on the Laboratory Evaluation and Assistance Check List.

a. Scheduling might necessitate starting the evaluation midway on the list of tasks for processing a sample, instead of at the beginning. In that case, the Evaluator will view the activities of the Sorter when a new sample is started to complete the evaluation of the first activities on the list.
b. If a Sorter or QC Officer misses or incorrectly performs a procedure, the Evaluator will note this on the checklist and *immediately point this out so the mistake can be corrected on the spot*. The role of the Evaluator is to provide additional training and guidance so that the procedures are being performed consistent with the *Benthic Laboratory Methods* manual, all data are recorded correctly, and paperwork is properly completed at the site.
c. When the sample has been completely processed, the Evaluator will review the results of the evaluation with the Sorter and QC Officer, noting positive practices and problems, weaknesses [might affect data quality], and deficiencies [would adversely affect data quality]). The Evaluator will ensure that the Sorter and QC Officer understand the findings and will be able to perform the procedures properly in the future.
d. The Evaluator will record responses or concerns, if any, on the Laboratory Evaluation and Assistance Check List.
e. If the Evaluator's findings indicate that Laboratory staff are not performing the procedures correctly, safely, or thoroughly, the Evaluator must continue working with these staff members until certain of their ability to process the sample properly so that data quality is not adversely affected.
f. If the Evaluator finds major deficiencies in the Laboratory operations, the Evaluator must contact one of the following QA officials:

Dr. Esther Peters, Tetra Tech QA Officer (703-385-6000)
Jennifer Hanson, GLEC QA Officer (231-941-2230)
Ms. Sarah Lehman, EPA NRSA Project QA Officer (202-566-1379)

The QA official will contact the Project Implementation Coordinator (Ellen Tarquinio – 202-566-2267) to determine what should be done.

Data records from samples previously processed by this Laboratory will be checked to determine whether any samples must be redone.

g. Complete the Laboratory Evaluation and Assistance Check List, including a brief summary of findings, and ensure that the Sorter and QC Officer have read this and signed off before leaving the laboratory.

9. The Evaluator will electronically scan and make a photocopy of the Laboratory Evaluation and Assistance Check List. The Evaluator will retain the photocopied checklist, and email the scanned file and send the original checklist to

Richard Mitchell
USEPA Office of Wetlands, Oceans, and Watersheds
1200 Pennsylvania Avenue (4503-T)
Washington, DC 20460-0001
(202)-566-0644
6.

## 7.0 REFERENCES

American Public Health Association. 1989. *Standard Methods for the Examination of Water and Wastewater.* Seventeenth Edition. American Public Health Association, Washington, D.C.

Bain, M.B., J.T. Finn, and H.E. Booke. 1985. Quantifying stream substrate for habitat analysis studies. North American Journal of Fisheries Management 5:499-500.

Baker, J.R. and G.D. Merritt, 1990. Environmental Monitoring and Assessment Program: Guidelines for Preparing Logistics Plans. EPA 600/4-91-001. U.S. Environmental Protection Agency. Las Vegas, Nevada.

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates, and Fish. Second Edition. EPA/841-B-99-002. U.S. Environmental Protection Agency, Office of Water, Assessment and Watershed Protection Division, Washington, D.C.

Bickford, C.A., C.E. Mayer, and K.D. Water. 1963. An Efficient Sampling Design for Forest Inventory: The Northeast Forest Resurvey. Journal of Forestry. 61: 826-833.

CAS. 1999. Chemical Abstracts Service web site (http://www.cas.org)

Carlson, R.E. 1977. A trophic state index for lakes. *Limnology and Oceanography* 22(2):361-369.

CENR. 1997. Integrating the Nation's Environmental Monitoring and Research Networks and Programs: A Proposed Framework. Committee on Environment and Natural Resources, National Science and Technology Council, Washington, DC, USA.

Chaloud, D.C., J.M. Nicholson, B.P. Baldigo, C.A. Hagley, and D.W. Sutton. 1989. *Handbook of Methods for Acid Deposition Studies: Field Methods for Surface Water Chemistry.* EPA 600/4-89/020. U.S. Environmental Protection Agency, Washington, D.C.

Converse, J.M. 1987. *Survey Research in the United States: Roots and Emergence 1890-1960*. University California Press. Berkeley, CA. 564 pp.

EIMS. 1999. Environmental Information Management System (EIMS) web site. (Http://www.epa.gov/eims)

FGDC. 1998. Content standard for digital deospatial metadata, version 2.0. FGDCSTD-001-1998. Federal Geographic Data Committee. Washington, DC. (Http://www.fgdc.gov)

Frissell, C.A., W.J. Liss, C.E. Warren, and M.D. Hurley. 1986. A hierarchical framework for stream habitat classification: viewing streams in a watershed context. *Environ. Mgmt.* 10(2): 199-214.

Frithsen, J.B. 1996a. Suggested modifications to the EMAP data set directory and catalog for implementation in US EPA Region 10,. Draft. June 10, 1996. Report prepared for the U.S. Environmental Protection Agency. National Center for Environmental Assessment. Washington, DC. By Versar, Inc., Columbia, MD.

Frithsen, J.B. 1996b. Directory Keywords: Restricted vs. unrestricted vocabulary. Draft, May, 21, 1996. Report prepared for the U.S. Environmental Protection Agency, National Center for Environmental Assessment, Washington, DC., by Versar, Inc. Columbia, MD.

Frithsen, J.B., and D.E. Strebel. 1995. Summary documentation for EMAP data: Guidelines for the information management directory. 30 April 1995. Report prepared for U.S. Environmental Protection Agency, Environmental and Assessment Program (EMAP), Washington, DC. Prepared by Versar, Inc., Columbia, MD.

Garner, F.C., M.A. Stapanian, and K.E. Fitzgerald. 1991. Finding causes of outliers in multivariate environmental data. Journal of Chemometrics. 5: 241-248.

Glase, J.A., D.L. Foerst, G.D> McKee, S.A. Quave, and W.L. Budde. 1981. Trace analyses of waste-waters. Environmental Science & Technology. 15: 1426-1435.

Hawkins, C. P., R. H. Norris, J. N. Hogue, and J. W. Feminella. 2000. Development and evaluation of predictive models for measuring the biological integrity of streams. *Ecological Applications* 10:1456-1477.

Hazard, J.W., and B.E. Law. 1989. Forest Survey Methods Used in the USDA Forest Service,. EPA/600/3-89/065. NTIS PB89 220 594/AS. U.S. EPA Environmental Research Laboratory. Corvallis, Oregon.

Hillman, D.C., S.H. Pia, and S.J. Simon. 1987. *National Surface Water Survey: Stream Survey (Pilot, Middle Atlantic Phase I, Southeast Screening, and Episode Pilot) Analytical Methods Manual.* EPA 600/8-87-005. U.S. Environmental Protection Agency, Las Vegas, Nevada.

Heinz Center. 2002. *The State of the Nation's Ecosystems.* The Cambridge University Press.

Hunsaker, C. T., and D. E. Carpenter. 1990. Environmental Monitoring and Assessment Program: ecological indicators. Office of Research and Development, U. S. Environmental Protection Agency, Research Triangle Park, North Carolina. EPA-600-3-90-060.

Hunt, D.T. E., and A.L. Wilson. 1986 The Chemical Analysis of Water: General Principles and Techniques. Second edition. Royal Society of Chemistry, London, England. 683 pp.

ITIS. 1999. Integrated Taxonomic Information System web site (http://www.itis.usda.gov/itis).

Kaufmann, P.R. (ed.). 1993. Physical Habitat. IN: R.M. Hughes (ed.) *Stream Indicator and*

*Design Workshop*. EPA600/R-93/138. U.S. Environmental Protection Agency, Corvallis, Oregon.

Kaufmann, P.R., A. T. Herlihy, J.W. Elwood, M.E. Mitch, W.S. Overton, M.J. Sale, J.J.. Messer, K.A. Cougan, D.V. Peck, K.H. Reckhow, A,J, Kinney, S.J. Christie, D.D. Brown, C.A. Hagley, and H.I. Jager. 1988. *Chemical Characteristics of Streams in the Mid-Atlantic and Southeastern United States. Volume I: Population Descriptions and Physico-Chemical Relationships.* EPA 600/3-88/021a. U.S. Environmental Protection Agency, Washington, D.C.

Kish, L. 1965. Survey Sampling. John Wiley & Sons. New York. 643 pp.

Kish, L. 1987. Statistical Design for Research. John Wiley & Sons. New York. 267 pp.

Kirchmer, C.J. 1983. Quality control in water analysis. Environmental Science & Technology. 17: 174A-181A.

Klemm, D.J., P.A. Lewis, F. Fulk, and J.M. Lazorchak. 1990. *Macroinvertebrate Field and Laboratory Methods for Evaluating the Biological Integrity of Surface Waters*. EPA 600/4-90/030. U.S. Environmental Protection Agency, Cincinnati, Ohio.

Lemmon, P.E. 1957. A new instrument for measuring forest overstory density. *J. For.* 55(9): 667-669.

Linsley, R.K., M.A. Kohler, and J.L.H. Paulhus. 1982. Hydrology for Engineers. McGraw-Hill Book Co. New York.

Meglen, R.R. 1985. A quality control protocol for the analytical laboratory. Pp. 250-270 IN: J.J. Breen and P.E. Robinson (eds). Environmental Applications of Cehmometrics. ACS Symposium Series 292. American Chemical Society, Washington, D.C.

Messer, J.J., C.W. Ariss, J.R. Baker, S.K. Drousé, K.N. Eshleman, P.R. Kaufmann, R.A. Linthurst, J.M. Omernik, W.S. Overton, M.J. Sale, R.D. Schonbrod, S.M. Stambaugh, and J.R. Tuschall, Jr. 1986. *National Surface Water Survey: National Stream Survey, Phase I-Pilot Survey.* EPA-600/4-86/026. Washington, D.C: U.S. Environmental Protection Agency.

MRLC. 1999. Multi-Resolution Land Characteristics web site (http://www.epa.gov/mrlc)

Mulvey, M., L. Cato, and R. Hafele. 1992. *Oregon Nonpoint Source Monitoring Protocols Stream Bioassessment Field Manual: For Macroinvertebrates and Habitat Assessment*. Oregon Department of Environmental Quality Laboratory Biomonitoring Section. Portland, Oregon. 40pp.

NAPA. 2002. *Environment.gov.* National Academy of Public Administration. ISBN: 1-57744-083-8. 219 pages.

NBII. 1999. The NBII Biological Metadata Standard. National Biological Information

Infrastructure web site (http://www.nbii.gov)

NSDI. 1999. National Spatial Data Infrastructure web site (http://www.fgdc.gov/nsdi/nsdi.html)

NRC. 2000. *Ecological Indicators for the Nation.* National Research Council.

Washington, DC. National Academy Press.

Overton, W.S., 1985. Draft Sampling Plan for Streams in the National Surface Water Survey. July 1986. *Technical Report 114*. Corvallis, Oregon: Department of Statistics, Oregon State University.

Overton, W. S., D. White, and D. L. Stevens, Jr. 1991. Design Report for EMAP, the Environmental Monitoring and Assessment Program. U. S. Environmental Protection Agency, Office of Research and Development, Washington, D.C. EPA-600-3- 91-053.

Paulsen, S.G., D.P. Larsen, P.R. Kaufmann, T.R. Whittier, J.R. Baker, D. Peck, J. McGue, R.M. Hughes, D. McMullen, D. Stevens, J.L. Stoddard, J. Lazorchak, W. Kinney, A.R. Selle, and R. Hjort. 1991. EMAP - surface waters monitoring and research strategy, fiscal year 1991. EPA-600-3-91-002. U.S. Environmental Protection Agency, Office of Research and Development, Washington, D.C. and Environmental Research Laboratory, Corvallis, Oregon.

Peck, D.V., J.M. Lazorchak, and D.J. Klemm (editors). 2003. Unpublished draft. Environmental Monitoring and Assessment Program – Surface Waters: Western Pilot Study Field Operations Manual for National Rivers and Streams. EPA/xxx/x-xx/xxxx. U.S. Environmental Protection Agency, Washington, D.C.

Plafkin, J.L., M.T. Barbour, K.D. Porter, S.K. Gross, and R.M. Hughes. 1989. *Rapid Bio-assessment Protocols for Use in Streams and Rivers: Benthic Macroinvertebrates and Fish.* EPA 440/4-89/001. U.S. Environmental Protection Agency, Office of Water, Washington, D.C.

Platts, W.S., W.F. Megahan, and G.W. Minshall. 1983. *Methods for Evaluating Stream, Riparian, and Biotic Conditions*. USDA Forest Service, Gen. Tech. Rep. INT-183. 71pp.

Robison, E.G. and R.L. Beschta. 1990. Characteristics of coarse woody debris for several coastal streams of southeast Alaska, USA. Canadian Journal of Fisheries and Aquatic Sciences 47(9):1684-1693.

Robison, E.G., and P.R. Kaufmann. (In preparation). Evaluating and improving and objective rapid technique for defining pools in small National Rivers and Streams.

Sarndal, C.E., B. Swensson, and J. Wretman. 1992. Model Assisted Survey Sampling. Springer-Verlag. New York. 694 pp.

Skougstad, M.W., M.J. Fishman, L.C. Friedman, D.E. Erdman, and S.S. Duncan (eds.). 1979.

Method I-4600-78, Automated Phosphomolybdate Colorimetric Method for Total Phosphorus. IN: *Methods for Determination of Inorganic Substances in Water and Fluvial Sediments: Techniques of Water-Resources Investigations of the United States Geological Survey*. Book 5, Chapter A1. U.S. Government Printing Office, Washington, D.C.

Smith, F., S. Kulkarni, L. E. Myers, and M. J. Messner. 1988. Evaluating and presenting quality assurance data. Pages 157-68 in L.H. Keith, ed. ACS Professional Reference Book. Principles of Environmental Sampling. American Chemical Society, Washington, D.C.

Stack, B.R. 1989. *Factors Influencing Pool Morphology in Oregon Coastal Streams*. M.S. Thesis, Oregon State University. 109pp.

Stanley, T.W., and S.S. Verner. 1986. The U.S. Environmental Protections Agency's quality assurance program. pp. 12-19 IN: J.K. Taylor and T.W. Stanley (eds.). Quality Assurance for Environmental Measurements. ASTM STP 867, American Society for Testing and Materials, Philadelphia, Pennsylvania.

Stapanian, M.A., F.C. Garner, K.E. Fitzgerald, G.T. Flatman, and J.M. Nocerino. 1993. Finding suspected causes of measurement error in multivariate environmental data. Journal of Chemometrics. 7: 165-176.

Stevens, D. L., Jr., 1994. Implementation of a National Monitoring Program. Journal Environ. Management 42:1-29.

Stevens, D.L., Jr., and A.R. Olsen. 1999. Spatially restricted surveys over time for aquatic resources. Journal of Agricultural, Biological and Environmental Statistics. 4:415-428.

Stevens, D.L., Jr., and A.R. Olsen. 2003. Variance estimation for spatially balanced samples of environmental resources. Environmetrics. 14:593-610.

Stevens, D.L., Jr., and A.R. Olsen. 2004. Spatially-balanced sampling of natural resources. Journal of American Statistical Association. 99:262-278.

STORET. 1999. The STORET web site. (Http://www.epa.gov/OWOW/STORET)

U.S. EPA. 1987. *Handbook of Methods for Acid Deposition Studies: Laboratory Analyses for Surface Water Chemistry.* EPA/600/4-87/026. U.S. Environmental Protection Agency, Office of Research and Development, Washington, D.C.

U.S. EPA. 1991. IRM Policy Manual. Chapter 13. Locational data.

U.S. EPA. 1996b. Addendum to: Guidelines for the information management directory. U.S. EPA, NHEERL. Atlantic Ecology Division. Narragansett, RI.

U.S. EPA. 2003. *Draft Report on the Environment*. ORD and OEI. EPA-260-R-02-006.

USGCRP. 1998. Data Management for Global Change Research. Policy Statements for the National Assessment Program. July 1998. U.S. Global Change Research Program. National Science Foundation. Washington, DC.

Wilen, B.O. 1990. The U.S. Fish and Wildlife Service's National Wetlands Inventory. in S. J. Kiraly, R. A. Cross, J. D. Buffington (Eds.) *Federal Coastal Wetlands Mapping Programs* (U.S. Department of the Interior Fish and Wildlife Service, Washington, D.C., 1990), FWS Biological Report 90(18), pp. 9-20.

Wolman, M.G. 1954. A method of sampling coarse river-bed material. Transactions of the American Geophysical Union 35(6):951-956.