United States Environmental Protection Agency
Office of Water
Office of Environmental Information
Washington, DC
EPA 843-R-10-003

National Wetland Condition Assessment

Quality Assurance Project Plan

Version 2, March 2012

QUALITY ASSURANCE PROJECT PLAN
REVIEW & DISTRIBUTION ACKNOWLEDGMENT AND
COMMITMENT TO IMPLEMENT

for

National Wetland Condition Assessment (NWCA)

We have read the QAPP and the methods manuals for NWCA listed below. Our agency/organization, agrees to abide by its requirements for work performed under NWCA (under CWA 106).

Quality Assurance Project Plan □
Field Operations Manual □
Site Evaluation Guidelines □
Laboratory Methods Manual □

Print Name _____

Title _____
(Cooperator's Principal Investigator)

Organization_____

_____ _____
Signature Date

NOTICE

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

- NWCA: Site Evaluation Guidelines (EPA-843-R-10-004)
- NWCA: Field Operations Manual (EPA-843-R-10-001)
- NWCA: Laboratory Methods Manual (EPA-843-R-10-002)
- Ecological Indicators for the 2011 National Wetland Condition Assessment (in preparation)

This document (Quality Assurance Project Plan) contains elements of the overall project management, data quality objectives, measurement and data acquisition, and information management for NWCA. Methods described in this document are to be used specifically in work relating to NWCA. All Project Cooperators should 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).

The suggested citation for this document is:

> USEPA. 2011 (draft). National Wetland Condition Assessment: Integrated Quality Assurance Project Plan. EPA 843-R-10-003. U.S. Environmental Protection Agency, Office of Water and Office of Research and Development, Washington, DC.

NWCA QAPP VERSION 2 SIGNATURE PAGE

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VERSION HISTORY

QAPP Version	Date Approved	Changes Made
1	4/26/2011	N/A
2	3/5/2012	Section 6.2

DISTRIBUTION LIST

This QA Project Plan and associated manuals or guidelines will be distributed to the following: EPA, States, Tribes, universities, and contractors participating in NWCA. EPA Regional Survey Coordinators are responsible for distributing NWCA QA Project Plan to State and Tribal Water Quality Agency staff or other cooperators who will perform the field sampling and laboratory operations. The Great Lakes Environmental Center 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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1.1 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 the U.S. Environmental Protection Agency (EPA), states, and tribes collectively 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, states, and tribes 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 and tribes 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?"

The most commonly cited and scientifically valid sources of national-scale wetland information are the U.S. Fish and Wildlife Service (FWS) *Wetlands Status and Trends Reports* (S&T Report), which have documented trends in wetland acreage since the 1950's. The most recent report, published in 2005, documented an annual net increase of 32,000 wetland acres from 1998-2004. At the same time, the report documents significant increases in freshwater ponds (12 percent) and alarming decreases in highly productive emergent mashes and coastal wetlands (Dahl 2005). In fact, a follow-up study recently published by the National Oceanic and Atmospheric Administration (NOAA) and U.S. FWS showed that wetlands in coastal watersheds of the eastern U.S. decreased at a rate of approximately 60,000 acres per year during the same study period (Stedman and Dahl, 2009). It is vitally important for wetland managers to understand the causes and sources of this loss to inform implementation of appropriate management measures. While the S&T Report is an invaluable source of information on trends in wetland acreage and class, it does not provide data on wetland condition.

In response to these needs, EPA Office of Water (OW), in concert with EPA's Office of Research and Development (ORD), the 10 EPA Regions, states and tribes has begun a program to assess the condition of the nation's waters via a statistically valid approach. The current assessment is the National Wetland Condition Assessment (referred to as NWCA). NWCA is a national assessment of the condition of the Nation's wetlands in the conterminous U.S. This assessment is the first assessment of wetlands to be based on data consistently collected using the same field and laboratory protocols, and based on a statistical survey design that allows inferences about all wetlands based on a sample of wetlands across the country. NWCA effort will provide important information about the condition of the nation's wetland resources and key stressors on a national and regional scale.

EPA developed this Quality Assurance Project Plan (QAPP) to support project participants 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 NWCA. EPA recognizes that states and tribes may add elements to the Survey, such as supplemental indicators, that are not covered in the

scope of this integrated QAPP. EPA expects that any supplemental elements are addressed by the states, tribes, or their designees, in a separate approved QAPP or an addendum to this QAPP. The NWCA participants have agreed to follow this QAPP and the protocols and design laid out in this document.

This cooperative effort between states, tribes, and federal agencies makes it possible to produce a broad-scale assessment of the condition of the Nation's wetlands with both confidence and scientific credibility. Development of the NWCA will build on the accomplishments of the USFWS and their production of national reports on status and trends in wetland acreage. When taken together, the NWCA and the S&T Report results will be used to measure progress toward attainment of the national goal to increase the quantity and quality of the Nation's wetlands. These complementary studies will influence how wetlands are managed at local, state, and national scales (Scozzafava, et. al. 2007).

USEPA will collaborate with state, tribal, federal, and other partners to implement the NWCA to meet three goals:

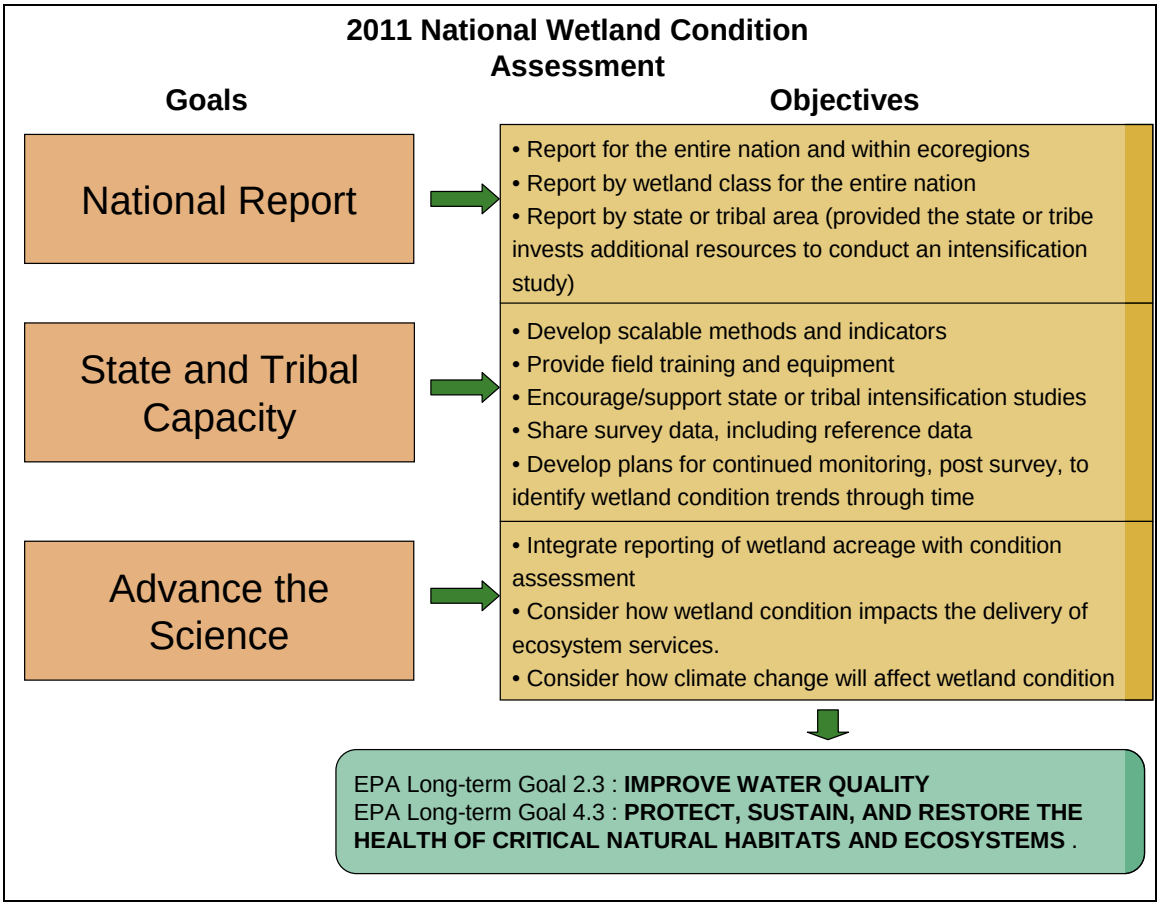
1. Produce a report that describes the ecological condition of the Nation's wetlands and ranks the predominant stressors associated with poor wetland condition.

2. Assist states and tribes in the implementation of wetland monitoring and assessment programs that will guide policy development and aid project decision-making.

3. Advance the science of wetlands monitoring and assessment to support management needs.

Through the framework of its goals and objectives, the NWCA addresses the long-term goals outlined in the Agency's current strategic plan (EPA 2006b) to improve the Nation's water quality and to protect, sustain, and restore the health of critical natural habitats and ecosystems, including wetlands (Figure 1-1).

Figure 1-1: **Relationship between the goals and objectives of the National Wetland Condition Assessment and the long-term goals of EPA's current strategic plan (EPA 2006b)**



1.2 NWCA Project Organization

A comprehensive quality assurance (QA) program, including assigning roles and responsibilities, was established to ensure data integrity and provide support for the reliable interpretation of the findings from this project. The responsibilities and accountability of the various principals and cooperators are described here and illustrated in Figure 1-2. The overall coordination of the project will be provided by EPA's Office of Water (OW) in Washington, DC, with support from EPA's Office of Research and Development (ORD). Each EPA Regional Office has identified a Regional EPA Coordinator to provide the critical link with state and tribal partners. State and Tribal Cooperators will work with their Regional EPA Coordinator to address any technical issues. In addition, the National Wetlands Monitoring and Assessment Work Group (NWMAWG) is a group of experts from the federal family, states, tribes, academia, and other cooperators. Subsets of the NWMAWG membership will be tasked to decide on the most appropriate approaches for key technical issues, such as: (1) the selection and establishment of reference conditions based on least-disturbed sites and expert consensus for characterizing benchmarks for assessment of ecological condition; (2) selection and calibration of ecological endpoints and attributes of the biota and relationship to stressor indicators; (3) a data analysis plan for interpreting the data and addressing the objectives in a nationwide assessment; and (4)

a framework for the reporting of the condition assessment and conveying the information on the ecological status of the Nation's wetlands.

Contractor support is provided for all aspects of this project. Contractors will provide support ranging from implementing the survey, sampling and laboratory processing, data management, data analysis, and report writing. Cooperators will interact with their Regional EPA Coordinator and the EPA Project Leader regarding contractual services.

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

Project Leader – Michael Scozzafava, OW

- 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.

Alternate Project Leaders – Chris Faulkner and Gregg Serenbetz, OW

- Assists EPA Project Leader with coordination and assumes responsibility for certain aspects of the project, as agreed upon with the EPA Project Leader.
- Serves as primary point-of-contact for project coordination in the absence or unavailability of Project Leader.
- Serves on the Technical Experts Workgroup and interacts with Project Leader on technical, logistical, and organizational issues on a regular basis.

QA Assistance Visit Coordinator – Regina Poeske, Region 3

- Assists in the implementation of the QA program.
- Coordinates all field and laboratory quality assurance visits.

EPA Project QA Lead – Sarah Lehman, OW

- Provides leadership, development and oversight of project-level quality assurance for NWCA in Office of Water

Technical Advisor – Mary Kentula, ORD (Teresa Magee, alternate)

- Advises the Project Leader on the relevant experiences and technology developed within ORD that are to be used in this project.
- Facilitates consultations between NWCA personnel and ORD scientists.

EPA (OWOW) QA Officer – Margarete Heber, OW

- Functions as the primary officer overseeing all QA and quality control (QC) activities.
- Responsible for ensuring that the QA program is implemented thoroughly and adequately to document the performance of all activities.

Regional EPA Coordinators

- Assist Project Leader with regional coordination activities.
- Serve on the NWMAWG and interact with Project Leader on technical, logistical, and organizational issues on a regular basis.

- Serve as primary points-of-contact for the Cooperators.

Contractor Technical Representative

- Provides contractor support to the project and works with Project Leader to ensure all needs for contractor support are covered.

Study Design Manager – Tony Olsen, ORD

- Coordinates w/ Project Lead, REPACs, NWMAWG, and Field Implementation Coordinator to develop and manage the Sampling Frame, select sampling locations, and track field evaluation and site reconnaissance.

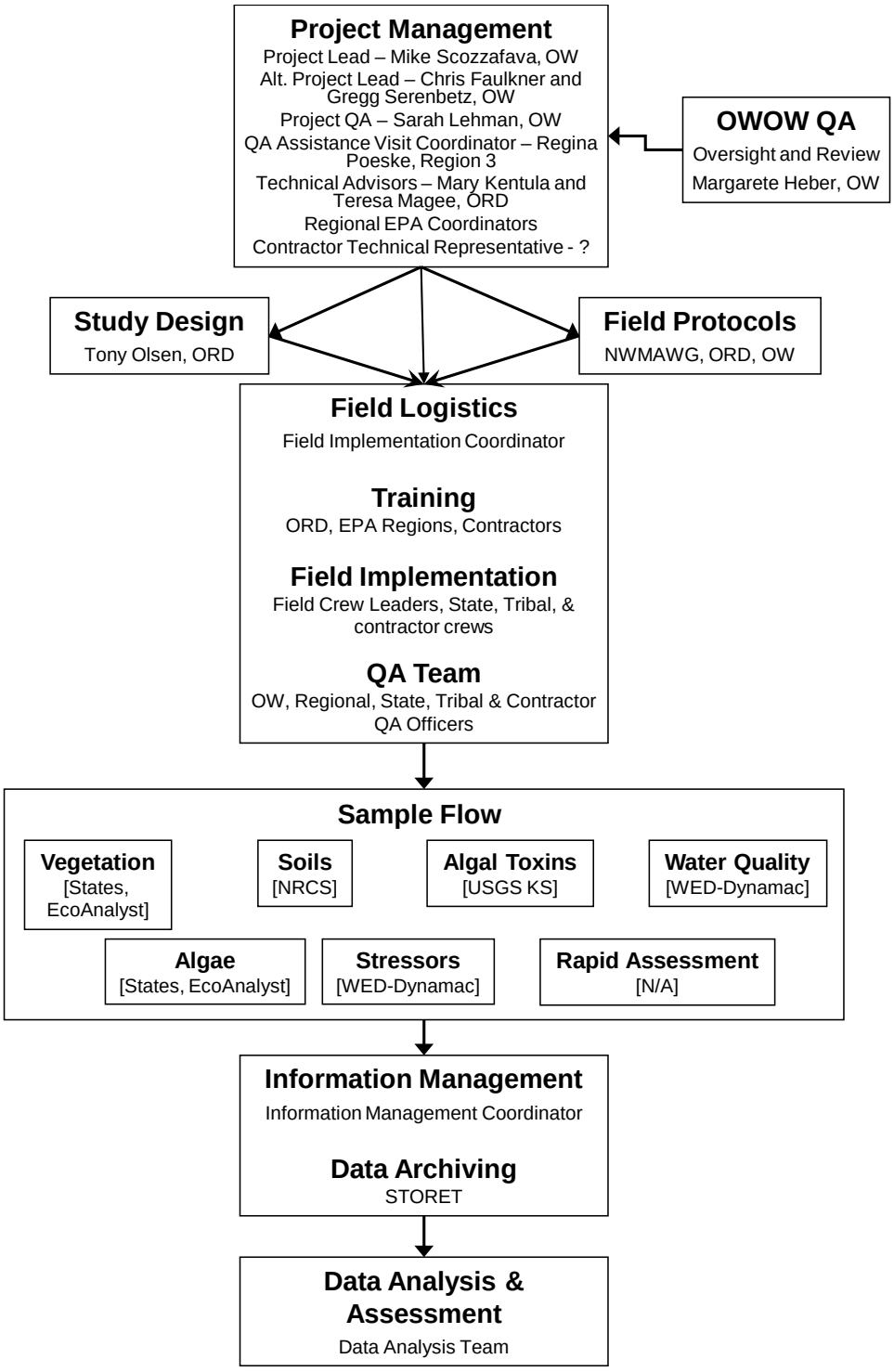


Figure 1-2**: NWCA Project Organization**

NWMAWG – States, EPA, academics, other federal agencies

- Provides expert consultation on key technical issues as identified by the EPA Project Management team and works with Project Lead to resolve approaches and strategies to enable data analysis and interpretation to be scientifically valid.

Logistics Coordinator – Dennis McCauley, Great Lakes Environmental Center (GLEC)

- A contractor who functions to support implementation of the project based on technical guidance established by the EPA Project Leader and Alternate EPA Project Leader serves as point-of-contact for questions from field crews and cooperators for all activities.
- Tracks progress of field sampling activities.
- Tracks progress of lab activities.

Field Crew Leader

- Functions as the senior member of each Cooperator's field sampling crew and the point of contact for the Field Implementation Coordinator.
- Responsible for overseeing all activities of the field sampling crew and ensuring that the Project field method protocols are followed during all sampling activities.

Cooperator(s) – States, Tribes, academics, other federal agencies, contractors

- Under the scope of their assistance agreements, plan and execute their individual studies as part of the cross jurisdictional NWCA, and adhere to all QA requirements and standard operating procedures (SOPs).
- Interact with the Regional EPA Coordinators, Field Implementation Coordinator and EPA Project Leader regarding technical, logistical, organizational issues.

QA Team

- Oversees the transfer of samples and related records for each indicator.
- Ensures the validity of data for each indicator.

Regional QA Project Officer(s)

- Oversee(s) individual studies of cooperators (assistance recipients).
- Interacts with EPA Project Leader and Field Implementation Coordinator on issues related to sampling design, project plan, and schedules for conduct of activities.
- Collects copies of all official field forms, field evaluation checklists and reports.
- Oversees and maintains records on field evaluation visits, but is not a part of sampling teams.

State QA Officer(s)

- Work(s) with EPA Project Lead, Regional EPA Coordinators, and Field Implementation Coordinator to ensure that the NWCA field protocols and QA protocols are carried out.

Contractor QA Officer(s)

- The contractor QA Officer who will supervise the implementation of the QA program.

- Directs the field and laboratory audits and ensures the field and lab auditors are adequately trained to correct errors immediately to avoid erroneous data and the eventual discarding of information from the assessment.

Information Management Coordinator – Marlys Cappaert, WED-SRA

- A contractor who functions to support implementation of the project based on technical guidance established by the EPA Project Leader and Alternate EPA Project Leader oversees all sample shipments and receives data forms from the Cooperators.
- Oversees all aspects of data entry and data management for the project.

Data Analysis Team – EPA OW, ORD, Regions, FWS, NRCS, States, Michigan State Univ, Kenyon College, Utah State Univ, Oregon State Univ, and contractors

- Develops and implements the NWCA Data Analysis plan which details our approach for analyzing acquired data, generating metadata, developing one or more IBIs, discussing findings with stakeholders in workshops and other venues, and contributing to the final report.

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

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 wetlands by no later than December, 2013. Tasks leading up to the final report are described throughout the QAPP.

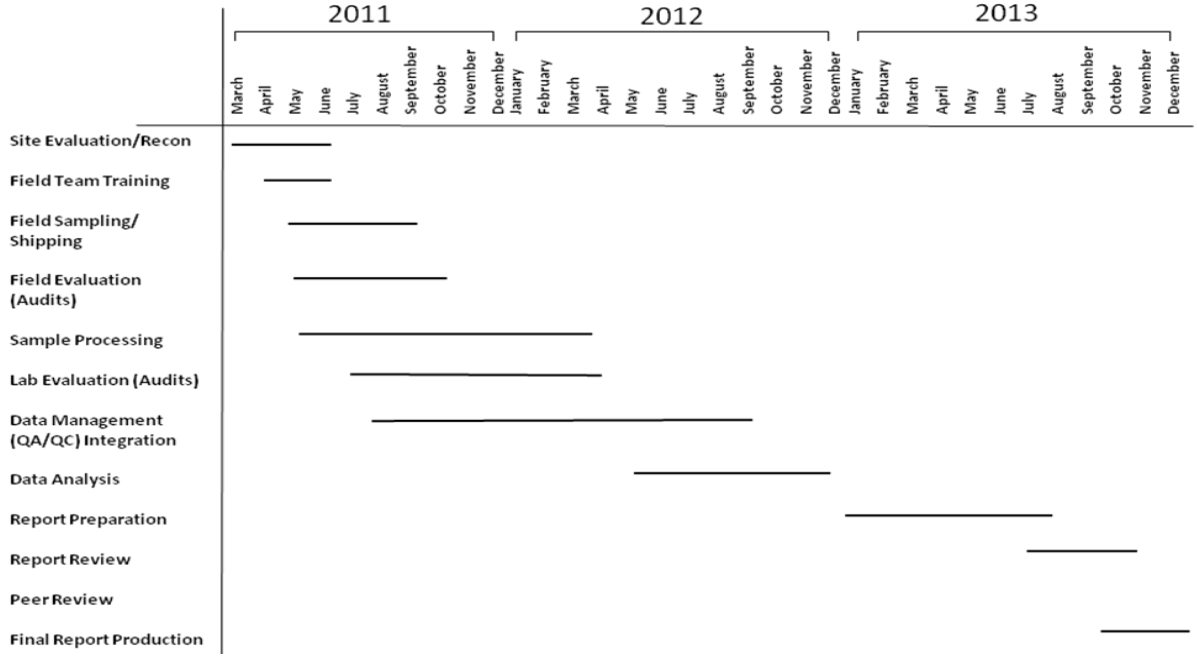


Figure 1-3: **Timeline of NWCA Activities**

1.3 Scope of QA Project Plan

This QA Project Plan addresses all aspects of the data acquisition efforts of the NWCA, which focuses on the 2011 sampling of wetland sites in the conterminous United States. Analysis of data from approximately 1000 sites (selected with a probability design) will provide a comprehensive assessment of the Nation's wetlands. Relevant companion documents to this QAPP include the following, which can be found at the NWCA web site (http://water.epa.gov/type/wetlands/assessment/survey/index.cfm):

- NWCA: Site Evaluation Guidelines (EPA 843-R-10-004)
- NWCA: Field Operations Manual (EPA 843-R-10-001)
- NWCA: Laboratory Methods Manual (EPA 843-R-10-002)
- Ecological Indicators for the 2011 National Wetland Condition Assessment (in preparation)

1.3.1 Overview of Field Operations

Field measurements and samples are collected by trained teams. Typically, each Field Crew is comprised of 4 members, divided into the Vegetation (Veg) Team and the Assessment Area and Buffer (AB) Team. The number and size of crews 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 two teams will work closely with each other, and coordinate sampling activities.

1.3.1.1 Field Crew Duties and Qualifications

The NWCA **Veg Team** is composed of a **Botanist/Ecologist** and a **Botanist Assistant**. Primary responsibilities for the Veg Team include:

1. Laying out the Assessment Area (AA) and vegetative plots;
2. Collecting high quality plant ecological data (including species identities, presence and cover of individual species, presence and cover of vertical vegetation strata, and counts of larger trees);
3. Collecting other information related to vegetation condition; and
4. Collecting and processing plant specimens.

The Veg Team carefully follows protocols to make onsite decisions regarding layout and set-up of the vegetation plots within the assessment area and to collect ecological data. Accurate plant species identification is critical to data quality. Careful descriptions of diagnostic characteristics, habitat, and plant associations will be documented. Plant specimens must be collected for all unknown taxa and quality assurance taxa, which will be later identified by expert taxonomists. Careful attention to providing tracking information for all specimens is essential.

In addition, NWCA will provide Veg Team members with training on study goals, vegetation sampling methods, field protocols, and plant collection requirements. Training will prepare the Team to accurately complete data and specimen collection tasks.

In addition to the skills developed in the training, the Botanist/Ecologist will have the following minimum qualifications:

- Understanding of basic wetland plant ecology.
- Familiarity with regional flora and proficiency in identifying common wetland plant species:
    - capable of sight recognition of often dominant species to the level of genus and species, provided plants are at the proper phonological stage; or
    - capable of sight recognition of dominant species to the family, and proficiency in keying in the field.
- Proficiency in keying many unknown plants (e.g., forbs, shrubs, trees) to species using regionally appropriate floras and diagnostic keys.
- Ability to distinguish difficult graminoid taxa as Poaceae (grasses), Juncaceae (rushes), and Cyperaceae (sedges, bulrushes, spikerushes), and to distinguish unknown species within these families or genera from one another.
- College course-work in plant taxonomy or systematics that included field identification of plant species; and/or excellent references regarding proficiency in botanical identification.
- Previous experience conducting botanical or ecological field work, including the collection and preservation of plant specimens.

All Botanist/Ecologist applicants will send their Curriculum vitae and references to the Regional Lead. The Project Lead and RMCs will review and verify the qualifications of all applicants prior to the applicants joining a Field Crew. If a State is unable to identify a Botanist/Ecologist, EPA will work with the State to identify a Botanist/Ecologist.

The NWCA **AB Team** is composed of two crew members, whose primary responsibilities include:

1. Collecting high-quality biological (e.g.,% vegetative cover, water quality), hydrology, soils and stressor data following the FOM and USA RAM protocols,
2. Collecting and processing soil, algae and water chemistry specimens.

Field Crew Training

Each Field Crew Leader and Botanist/Ecologist must be trained at an EPA-sponsored training session prior to the start of the field season, along with as many crew members as possible. 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. Project organizations responsible for training oversight are identified in Figure 1-2. Training documentation will be maintained by the Project QA Officers.

The AB team carefully follows protocols in both FOM and USA RAM to make onsite decisions regarding the collection of ecological data and placement of soil pits. All samples (algae, soil, water chemistry) must be carefully collected, preserved, packed and catalogued for tracking.

AB Team members should have the following skills/abilities:

- Some previous experience conducting ecological field work;
- Ability to recognize evidence of human (or natural) landscape disturbance from the present or recent past.
- Ability to use common field equipment (compass, GPS, laser rangefinder, stadia rods, etc.).
- Experience measuring basic physical characteristics of soil,
- Knowledge of regional hydric soil indicators
- Knowledge of hydrogeomorphic classification

In addition, NWCA will provide AB Team members with additional training on study goals, biological and physical sampling methods, field protocols, and soil collection requirements. Training will prepare the Team to accurately complete data and tracking tasks.

1.3.1.2Field Operations Timeline

Field data acquisition activities are implemented for the NWCA (Table 1), based on guidance developed for earlier EMAP studies (Baker and Merritt 1990).

Table 1.3-1. Critical logistics elements (from Baker and Merritt, 1990)

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

Pre-Field Visit Activities

Survey preparation is initiated with selection of the sampling locations by EPA's Office of Research and Development (WED in Corvallis). The list of sampling locations is distributed to the EPA Regional Wetland Monitoring Coordinators and cooperators. With the sampling location list, State and Tribal cooperators can decide to what level they wish to participate (vs. requesting in-kind assistance). Participating State and Tribal Field Crews can then begin site reconnaissance on both the primary sites and alternate/replacement sites (known as *base* and *oversample* locations, respectively) and begin work on obtaining access permission to each site[1].

Field Crews need to acquire permission to access sites on private property, as well as permits to access and sample federally protected land. The Field Crew Leader should begin contacting private property owners (and the appropriate federal agency in the case of federally protected land) as early as 2010. As the design requires repeat visits to selected sites (i.e. for sampling), it is important for the Field Crews 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. More details on the timing and acquisition of property access permissions and permits are found in the *NWCA: Site Evaluation Guidelines* (USEPA 2010[a]).

Equipment Use During NWCA Field Activities

The timely receipt, proper use (including inspection and calibration), and maintenance of appropriate equipment are important contributors to acquiring quality data.

The Field Crews will use standard field equipment and supplies which are provided by EPA and contractors. The Field Implementation Coordinator will work with Regional Wetland Monitoring Coordinators, Cooperators, States, Tribes and Contractors to make certain the Field Crews have the required equipment and supplies in a timely fashion. Detailed lists of equipment required for each field protocol are contained in the *NWCA: Field Operations Manual* (USEPA 2010[b]).

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

The Field Crews will be responsible for the inspection, maintenance, and calibration of the equipment they use. Detailed information (including guidance) on equipment inspection, maintenance, and calibration, are contained in the *NWCA: Field Operations Manual* (USEPA 2010[b]).

In addition to the initial list of base and oversample sampling locations, Cooperators conducting field operations (i.e., States and Tribes that decide to conduct field operations themselves, and contractors performing in-kind support) will also receive and develop Site Packets for the base locations. Each Site Packet will contain the following applicable information:

- topographic and aerial maps with the POINT location (lat/long) marked,
- copies of written access permissions,

[1] Specific procedures for evaluating each sampling location and for replacing target sites are documented in the *NWCA: Site Evaluation Guidelines* (USEPA, 2010[a]).

- scientific collection permits,
- information brochures on the program for interested land owners,
- road maps, and local area emergency numbers.

Field Visit Activities

The site verification process is shown in Figure 1-4. Upon arrival at a site, the POINT location is verified by a Global Positioning System (GPS) receiver. Samples and measurements for various indicators are collected in a specified order (Figure 1-5). This order has been set up to minimize the impact of sampling for one indicator upon subsequent indicators; for example, vegetation sampling is to be completed before soil pits are dug and sampled. All methods are fully documented in step-by-step procedures in the *NWCA: Field Operations Manual* (USEPA 2011[b]). 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.

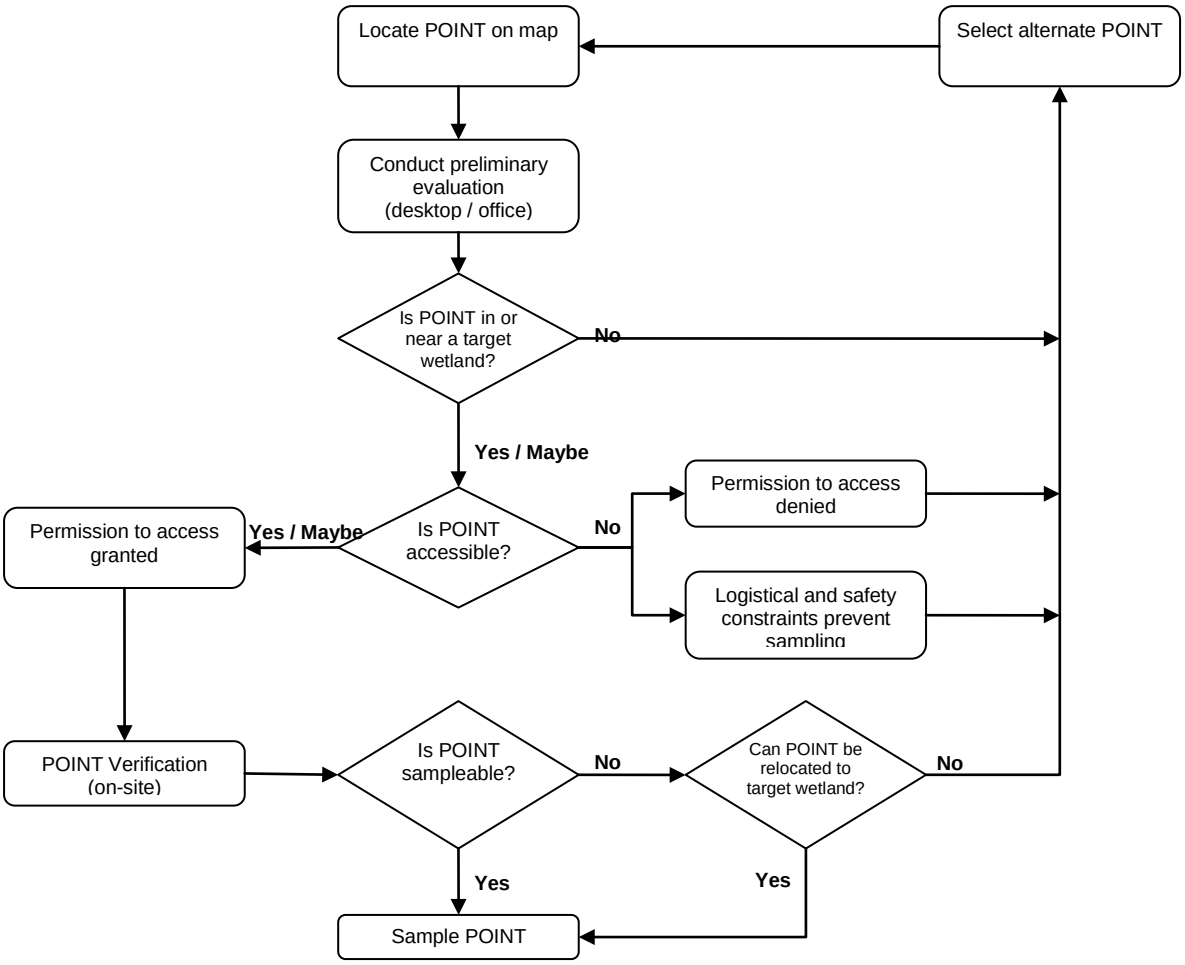


Figure 1-4: Site verification activities for wetland field surveys

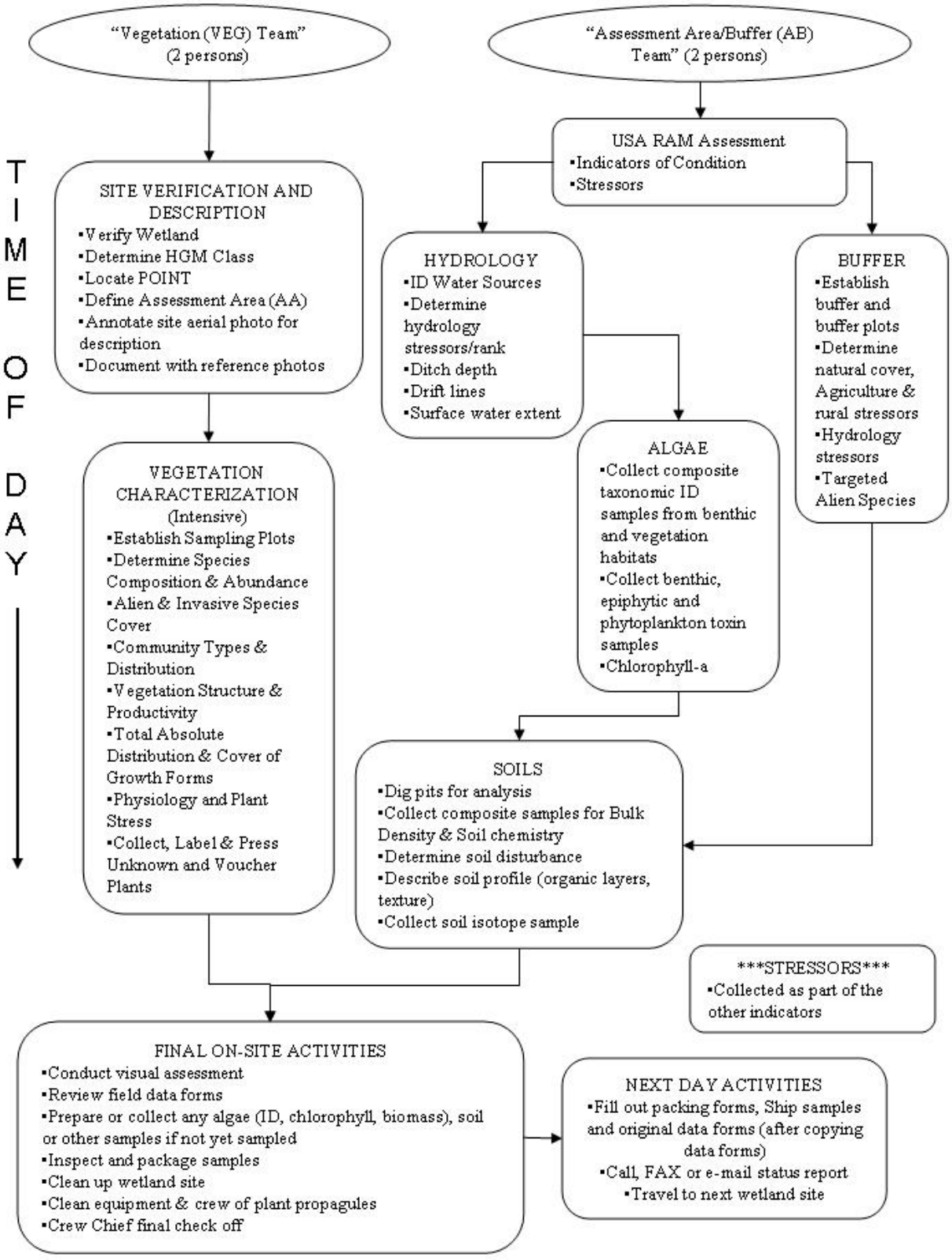


Figure 1-5*:* **Summary of field activities and site sampling**

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 Crew 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 provided by the design. All samples from a site must be labeled with this unique identifier.

Post-Field Visit Activities
Upon return from a field sampling site (either to the Field Crew'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 (refer to section 4.1.4 of this document for more information).

Samples are stored or packaged for shipment in accordance with instructions contained in the Field Operations Manual (USEPA 2010[b]). Samples which must be shipped are delivered to a commercial carrier. The recipient is notified to expect delivery; thus, tracing procedures can be initiated quickly in the event samples are not received. Bills of lading 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 Logistics 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). 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 analyses begin during sampling (e.g., *in situ* profiles) while others are not even initiated until sampling has been completed (e.g., algae). Analytical methods conducted in the field are described in the Field Operations Manual, and methods conducted in the laboratory are described in the Laboratory Methods Manual (USEPA, 2010[c]).

Chemical, physical, or biological analyses may be performed in-house or by contractor or cooperator laboratories. 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 are expected to conduct operations using good laboratory practices. General guidelines for analytical support laboratories:

- A program of scheduled maintenance of analytical balances, water purification systems, microscopes, laboratory equipment, and instrumentation.

- Verification of the calibration of analytical balances using class "S" weights which are certified by the National Institute of Standards and Technology (NIST).

- Verification of the calibration of top-loading balances using NIST-certified class "P" weights.

- Checking and recording the composition of fresh calibration standards against the previous lot. Acceptable comparisons are ≤ 2 percent of the theoretical value. (This acceptance is tighter than the method calibration criteria.)

- Recording all analytical data in bound logbooks in ink, or on standardized recording forms.

- Verification of the calibration of uniquely identified daily use thermometers using NIST-certified thermometers.

- Monitoring and recording (in a logbook or on a recording form) temperatures and performance of cold storage areas and freezer units (where samples, reagents, and standards may be stored). During periods of sample collection operations, monitoring must be done on a daily basis.

- An overall program of laboratory health and safety including periodic inspection and verification of presence and adequacy of first aid and spill kits; verification of presence and performance of safety showers, eyewash stations, and fume hoods; sufficiently exhausted reagent storage units, where applicable; available chemical and hazardous materials inventory; and accessible material safety data sheets for all required materials.

- An overall program of hazardous waste management and minimization, and evidence of proper waste handling and disposal procedures (90-day storage, manifested waste streams, etc.).

- If needed, having a source of reagent water meeting American Society of Testing and Materials (ASTM) Type I specifications for conductivity (< 1 µS/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 upon expiration.

- 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.

All laboratories providing analytical support to the NWCA must adhere to the provisions of this integrated QAPP. Laboratories will provide information documenting their ability to conduct the analyses with the required level of data quality before analyses begin. The documentation will

be sent to the EPA Project QA Lead (Sarah Lehmann) at EPA Headquarters. Such information might include results from inter-laboratory 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 Data Management Plan. Laboratory operations may be evaluated by technical systems audits, performance evaluation studies, and by participation in inter-laboratory sample exchange.

Table 1.3-2. 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 µS/cm at 25 ºC; 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 the EPA Project Leader is responsible for developing a data analysis plan that includes verification and validation. These processes are described in the internal indicator research strategies and summarized in the indicator-specific sections of this QAPP. Validated data are transferred to the central National Aquatic Resource Surveys (NARS) surface waters information management system at WED-Corvallis and managed by Information Management Staff. Data analysis to support this report will be conducted by the NWCA Data Analysis Team. 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. All validated measurement and indicator data from the NWCA will eventually be transferred to EPA's Water Quality Exchange (WQX) that will replace the STORET data management system.

1.3.4 Peer Review

The Survey will undergo a thorough peer review process, where the scientific community and the public will be given the opportunity to provide comments. Cooperators have been actively involved in the development of the overall project management, design, methods, and standards including the drafting of five key project documents:

- NWCA: Quality Assurance Project Plan (EPA 843-R-10-003)
- NWCA: Site Evaluation Guidelines (EPA 843-R-10-004)
- NWCA: Field Operations Manual (EPA 843-R-10-001)
- NWCA: Laboratory Methods Manual (EPA 843-R-10-002)
- Ecological Indicators for the 2011 National Wetland Condition Assessment (in preparation)

Outside scientific experts from universities, research centers, and other federal agencies have been instrumental in indicator development and will continue to play an important role in data analysis.

The EPA will utilize a three-tiered approach for peer review of the Survey: (1) internal and external review by EPA, states, other cooperators and partners, (2) external scientific peer review, and (3) public review.

Once data analysis is complete, cooperators will examine the results at regional meetings. Comments and feedback from the cooperators will be incorporated into the draft report. Public and scientific peer review will happen simultaneously. This public comment period is important to the process and will allow us to garner a broader perspective in examining the results before the final report. The public peer review is consistent with the Agency and OMB's revised requirements for peer review.

Below are the proposed measures EPA will implement for engaging in the peer review process:

1. Develop and maintain a public website with links to standard operating procedures, quality assurance documents, fact sheets, cooperator feedback, and final report
2. Conduct technical workgroup meetings composed of scientific experts, cooperators, and EPA to evaluate and recommend data analysis options and indicators
3. Hold national meeting where cooperators will provide input and guidance on data presentation and an approach for data analysis
4. Complete data validation on all chemical, physical and biological data
5. Conduct final data analysis with workgroup to generate assessment results
6. Engage peer review contractor to identify external peer review panel
7. Develop draft report presenting assessment results
8. Conduct regional meetings with cooperators to examine and comment on results
9. Develop final draft report incorporating input from cooperators and results from data analysis group to be distributed for peer and public review

10. Issue Federal Register (FR) Notice announcing document availability and hold scientific/peer review and public comment (30-45 days)
11. Consider scientific and public comments and produce a final report

The proposed peer review schedule is provided below and is contingent upon timeliness of data validation, schedule availability for regional meetings and experts for data analysis workshop.

May 2011 - December 2011	Data validation
March 15, 2012	Data analysis workshop
May - August 2013	Internal peer review meetings with states, cooperators, participants
October 19, 2013	Release for external peer and public review of draft

2 DATA QUALITY OBJECTIVES

It is U.S. EPA policy that Data Quality Objectives (DQOs) be developed for all environmental data collection activities following the prescribed DQO Process. DQOs are qualitative and quantitative statements that:

- Clarify study objectives;
- Define the appropriate types of data; and
- Specify the tolerable levels of potential decision errors.

These statements are the basis for establishing the quality and quantity of data needed to support decisions (EPA 2006). 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.

DQOs are typically expressed in terms of acceptable uncertainty (e.g., width of an uncertainty band or interval) associated with a point estimate at a desired level of statistical confidence (EPA 2006). The DQO Process is used to establish performance or acceptance criteria, which serve as the basis for designing a plan for collecting data of sufficient quality and quantity to support the goals of a study (EPA 2006). As a general rule, performance criteria represent the full set of specifications that are needed to design a data or information collection effort such that, when implemented, generate newly-collected data that are of sufficient quality and quantity to address the project's goals (EPA 2006). Acceptance criteria are specifications for evaluating the adequacy of existing sources of information or data as being acceptable to support the project's intended use (EPA 2006).

2.1 Data Quality Objectives for the National Wetland Condition Survey

Target DQOs established for the NWCA relate to the goal of describing the current status in the condition of selected indicators of the condition of wetlands in the conterminous U.S. and ecoregions of interest.

The formal statement of the DQO for national estimates is as follows:

> Estimate the proportion of wetlands (± 5%) in the conterminous U.S. that fall below the designated threshold for good conditions for selected measures with 95% confidence.

For the ecoregions of interest the DQO is:

> Estimate the proportion of wetlands (± 15%) in a specific ecoregion that fall below the designated threshold for good conditions for selected measures with 95% confidence.

2.2 Measurement Quality Objectives

For each parameter, performance objectives (associated primarily with measurement error) are established for several different data quality indicators (following USEPA [2002a]). Specific measurement quality objectives (MQOs) for each parameter are presented in the indicator section of this QAPP. The following sections define the data quality indicators and present approaches for evaluating them against acceptance criteria established for the program.

2.2.1 Laboratory Reporting Level (Sensitivity)

Generally, NWCA laboratory analyses can be divided between taxonomic (i.e. vegetation and algae, addressed in section 2.2.3) and non-taxonomic metrics. Non-taxonomic metrics are further broken out into physical metrics and chemical metrics.

For physical and chemical measurements, requirements for the method detection limit (MDL) are typically established. The MDL is defined as the lowest level of analyte that can be distinguished from zero with 99 percent confidence based on a single measurement (Glaser et al., 1981). USGS NWQL has developed a variant of the MDL called the long-term MDL (LT-MDL) to capture greater method variability (Oblinger Childress et al. 1999). Unlike MDL, it is designed to incorporate more of the measurement variability that is typical for routine analyses in a production laboratory, such as multiple instruments, operators, calibrations, and sample preparation events (Oblinger Childress et al. 1999). Because the LT-MDL addresses more potential sources of variability than the MDL, the NWCA uses the LT-MDL.

The LT-MDL determination ideally employs at least 24 blanks and spiked samples prepared and analyzed by multiple analysts on multiple instruments over a 6- to 12-month period at a frequency of about two samples per month (EPA 2004). The LT-MDL uses “F-pseudosigma” (F_σ) in place of s, the sample standard deviation, used in the EPA MDL calculation. F-pseudosigma is a non-parametric measure of variability that is based on the interquartile range of the data (EPA 2004). The LT-MDL is calculated using either the mean or median of a set of long-term blanks, and from long-term spiked sample results (depending on the analyte and specific analytical method). The LT-MDL for an individual analyte is calculated as:

Equation 1a

$$LT - MDL = M + \left(t_{0.99,n-1} \times F_{\sigma}\right)$$

where:

M = the mean or median of blank results
n = the number of spiked sample results
F_{σ} = F-pseudosigma, a nonparametric estimate of variability calculated as:

Equation 1b

$$F_{\sigma} = \frac{Q_3 - Q_1}{1.349}$$

where:

Q_3 = the 75th percentile of spiked sample results
Q_1 = the 25th percentile of spiked sample results

LT-MDL is designed to be used in conjunction with a laboratory reporting level (LRL; Oblinger Childress et al. 1999). The LRL is designed to achieve a risk of ≤1% for both false negatives and false positives (Oblinger Childress et al. 1999). The LRL is set as a multiple of the LT-MDL, and is calculated as follows:

LRL = (2 x LT–MDL)/fractional spike recovery

Where fractional spike recovery is the mean or median recovered spike concentration divided by the expected spike concentration. For example, at 50% recovery, LRL is 4 times the LT-MDL.
Therefore, multiple measurements of a sample having a true concentration at the LRL should result in the concentration being detected and reported 99 percent of the time (Oblinger Childress et al. 1999).

All laboratories will develop calibration curves for each batch of samples that include a calibration standard with an analyte concentration equal to the LRL. Estimates of LRLs (and how they are determined) are required to be submitted with analytical results. Analytical results associated with LRLs that exceed the objectives are flagged as being associated with unacceptable LRLs. Analytical data that are below the estimated LRLs are reported, but are flagged as being below the LRLs.

2.2.2 Sampling Precision, Bias, and Accuracy

Accuracy is a qualitative term referring to the proximity of a measurement to its "true" value. Accuracy will be qualitatively evaluated for taxonomic data collected as part of the NWCA, as described in Section 2.2.3 below; however, it will not be evaluated for all data. Precision and bias, on the other hand, are quantitative terms referring to the agreement between multiple measurements and the distance between those measurements and the true value (respectively). Precision and bias are estimates of random and systematic error in a measurement process (Kirchmer, 1983; Hunt and Wilson, 1986, USEPA 2002a). Collectively, precision and bias provide an estimate of the total error or uncertainty associated with an individual measurement or set of measurements. Precision and bias MQOs are developed for lab measurements. Precision, bias, and accuracy of field measurements will not be monitored during the NWCA[2].

[2] Bias, for example, cannot be determined directly, since the "true" values at any particular site are not known.

Laboratory Measurements

Systematic errors in water and soil chemistry metrics are minimized by using validated methods and standardized procedures across all laboratories. 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, MQOs for precision and bias are established in both absolute and relative terms, following the approach outlined in Hunt and Wilson (1986). At lower concentrations, MQOs are specified in absolute terms. At higher concentrations, MQOs are stated in relative terms. The point of transition between an absolute and relative MQO 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%).

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

Equation 1

$$RPD = \left(\frac{|A - B|}{(A + B)/2} \right) \times 100$$

Where:

A = the first measured value
B = the second measured value.

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

Equation 2

$$B[\%] = \frac{\bar{x} - T}{T} \times 100$$

Where:

x = the mean value for the set of measurements
T = the theoretical or target value of a performance evaluation sample.

Precision and bias within each laboratory are monitored for every sample batch by the analysis of internal QC samples. Samples associated with unacceptable QC sample results are reviewed and re-analyzed if necessary. Precision and bias across all laboratories will be evaluated after analyses are completed by using the results of performance evaluation (PE) samples sent to all laboratories (3 sets of 3 PE samples, with each set consisting of a low, moderate, and high concentration sample of all analytes).

Field Measurements

Since precision, bias, and accuracy of field measurements will not be monitored during the NWCA, a revisit site approach will be taken to ensure the quality of data. The survey design incorporates a plan for repeated sampling of a subset of sites. Data from these repeat visits provide estimates of important components of variance to evaluate the performance of ecological indicators. These variance components are presented in Table 2.2-1. If estimates of these components are available from other studies, they are used in conjunction with the project requirements to evaluate alternative design scenarios (Larsen et al. 1995, 2001, 2004). Status estimates are influenced most by the interaction (if multiple years are required to complete sampling) and residual variance components. Residual variance is composed of temporal variance within a sampling period confounded with measurement error of various types. If the

magnitude of residual variance is sufficiently large to impact status estimates (see above), then relative magnitudes of the interaction variance and various components of residual variance are examined to determine if any reduction can be achieved in the future. Interaction variance can only be reduced by increasing the sample size. Index variance can be reduced by either increasing the number of sites, increasing the number of times a site is visited within a year, reducing the length of the index period, or by reducing measurement error. Trend detection is evaluated using the equation to determine the variance in the slope of the trend (Table 2.2-1). In this model, residual variance also includes the interaction component. For multi-site networks such as the national aquatic resource assessments, trend detection is most sensitive to coherent year variance, which can only be reduced by extending the time period for monitoring (Larsen et al. 1995, 2001, 2004). If residual variance is large relative to the coherent year variance, then trend detection within a fixed time period can be improved by increasing the number of sites sampled each year, increasing the number of times each site is sampled within a year, or by reducing measurement error.

Table 2.2-1. Important variance components for aquatic resource assessments

Model for status estimation

$$\sigma^2_{total} = \sigma^2_{sites} + \left(\sigma^2_{year} + \sigma^2_{site\times year} + \sigma^2_{residual}\right)$$

and

$$\sigma^2_{residual} = \sigma^2_{within\text{-}year} + \sigma^2_{error}$$

Model for trend detection

$$\mathrm{var}(slope) = \frac{\frac{\sigma^2_{sites}}{N_{sites}} + \left(\sigma^2_{year} + \frac{s^2_{residual}}{N_{sites}}\right)}{\sum_{i=1}^{years}\left(y_i - \bar{Y}\right)^2}$$

and

$$s^2_{residual} = \sigma^2_{site\times year} + \frac{\sigma^2_{residual}}{N_{visit}}$$

Components in parentheses represent “extraneous” variance

Variance Component	Description
σ^2_{sites}	Observed variance among all sites or streams sampled over multiple-year sampling cycle. If sites are revisited across years, this effect can be eliminated
σ^2_{year}	Coherent variance across years that affects all sites equally, due to regional-scale factors such as climate or hydrology Principal effect on trend detection, reduced only by increasing number of years
$\sigma^2_{site\times year}$	“Interaction” variance occurring at each site across years that affects each site independently. Principal effect on status, reduce by increasing number of sites
$\sigma^2_{residual}$	“Residual” variance: Includes temporal variance at each site within a single index period ($\sigma^2_{within\text{-}year}$) confounded with measurement error (σ^2_{error}) due to acquiring the data from the site (e.g., sample collection and analysis) Principal effect on status, If $\sigma^2_{index} >> \sigma^2_{error}$ reduce by increasing number of sites or altering index period If σ^2_{error} is large relative to σ^2_{index}, then modify sampling and analysis procedures

For the NWCA ten percent of all sample sites will receive repeat visits to determine if differences exist in field data collection on different days. Revisit sites must be sampled at least 2-4 weeks apart to ensure that we are assessing temporal variability. Control measures to

minimize measurement error among crews and sites will be employed. These control measures include the use of standardized field protocols provided in the Field Operations Manual (FOM), consistent training of all crews, field assistance visits to all crews, and availability of experienced technical personnel during the field season to respond to site-specific questions from field crews as they arise.

Each Field Crew Leader and Botanist/Ecologist must be trained at an EPA-sponsored training session prior to the start of the field season, along with as many crew members as possible. 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 over the course of 3.5 days. Project organizations responsible for training oversight are identified in Figure 1-2. Training documentation will be maintained by the Project QA Officers.

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 to minimize data loss due to improper technique or interpretation of program guidance. The field visits evaluations will be based on the uniform training, plans, and checklists. For more information on field assistance visits see Chapter 6 of this document.

2.2.3 Taxonomic Precision and Accuracy

For the NWCA, 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 for vascular plants and algae, 10 percent of the samples will be randomly-selected for re-identification by an independent, outside taxonomist or laboratory. Comparison of the results of whole sample re-identifications will provide a Percent Taxonomic Disagreement (PTD) calculated as:

Equation 7

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

Where:

$comp_{pos}$ = the number of agreements
N = the total number of individuals in the larger of the two counts.

The lower the PTD, the more similar taxonomic results are and the overall taxonomic precision is better. A MQO of 15% is recommended for taxonomic differences (overall mean <15% is acceptable). Individual samples exceeding 15% are examined for taxonomic areas of substantial disagreement, and the reasons for disagreement investigated.

Where re-identification by an independent, outside taxonomist or laboratory is not practical (i.e., phytoplankton, algae), percent similarity will be calculated. Percent similarity is a measure of similarity between two communities or two samples (Washington 1984). Values range from 0% for samples with no species in common, to 100% for samples which are identical. It is calculated as follows:

Equation 8

$$PSC = 1 - 0.5\sum_{i=1}^{K}|a - b|$$

where:

a and b = for a given species, the relative proportions of the total samples A and B, respectively, which that species represents.

A MQO of ≥85% is recommended for percent similarity of taxonomic identification. If the MQO is not met, the reasons for the discrepancies between analysts should be discussed. If a major discrepancy is found in how the two analysts have been identifying organisms, the last batch of samples that have been counted by the analyst under review may have to be recounted.

Additionally, percent similarity should be calculated for re-processed subsamples. This provides a quantifiable measure of the precision of subsampling procedures employed for various parameters (i.e., phytoplankton, algae). A MQO of ≥70% is recommended for percent similarity of subsamples. If a sample does not meet this threshold, additional subsamples should be processed from that sample until the MQO is achieved.

Sample enumeration is another component of 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:

Equation 9

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

An MQO of 5% is recommended (overall mean of ≤5% is acceptable). Individual samples exceeding 5% are examined to determine reasons for the exceedance.

Corrective actions for samples exceeding these MQOs can include defining the taxa for which re-identification may be necessary (potentially even by a 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.

Taxonomic accuracy is evaluated by having individual specimens representative of selected taxa identified by recognized experts. Samples will be identified using the most appropriate technical literature that is accepted by the taxonomic discipline and reflects the accepted nomenclature. Where necessary, the Integrated Taxonomic Information System (ITIS, http://www.itis.usda.gov/) will be used to verify nomenclatural validity and spelling. A reference collection will be compiled as the samples are identified.

2.2.4 Completeness

Completeness requirements are established and evaluated from two perspectives. First, valid data for individual parameters 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 acquire valid data at 95% or more of the sampled sites. Percent completeness is calculated as:

Equation 10

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

Where:

V = the number of measurements/samples judged valid
T = the 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.

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 revisit samples (10% of sites visited) 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.

2.2.5 Comparability

Comparability is defined as the confidence with which one data set can be compared to another (USEPA 2002). A performance-based methods approach is being utilized for water chemistry analyses that define a set of laboratory method performance requirements for data quality. Following this approach, participating laboratories may choose which analytical methods they will use for each target analyte as long as they are able to achieve the performance requirements as listed in Table 5.4-1. For all parameters, comparability is addressed by the use of standardized sampling procedures and analytical methods by all sampling crews and laboratories. Comparability of data within and among parameters 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 NWCA contract laboratories.

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" (USEPA 2002). At one level, representativeness is affected by problems in any or all of the other data quality indicators.

At another level, representativeness is affected by the selection of the target wetlands, the location of sampling sites within that wetland, the time period when samples are collected, and the time period when samples are analyzed. The probability-based sampling design should estimate the condition of wetland 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 response design, (which includes when, where, and how to collect a sample at each site). Holding-time requirements for analyses ensure analytical results are representative of conditions at the time of sampling.

3 SAMPLING DESIGN AND SITE SELECTION

The overall sampling program for the National Wetland Condition Assessment project requires a randomized, probability-based approach for selecting wetlands where sampling activities are to be conducted. Details regarding the specific application of the probability design to surface waters resources are described in Paulsen et al. (1991) and Stevens (1994). The specific details for the collection of samples associated with different indicators are described in the indicator-specific sections of this QAPP.

3.1 Probability-Bases Sampling Design and Site Selection

The objectives, or design requirements, for the National Wetland Condition Assessment are to produce:

1. Estimates of the 2011 status of wetlands nationally and regionally (9 aggregated Omernik ecoregions, major river basins, EPA Regions, etc.),
2. Estimates of the 2011 status of seven S&T wetland classes nationally.
3. Estimates of the 2011 status of wetlands in coastal watersheds nationally,

Generally, almost all wetlands in the conterminous U.S. are considered the *target population* for the assessment (see the “Target population” sidebar for a more complete definition).

As stated in Chapter 1 section 1.1 (Introduction), the USFWS National Wetlands Inventory (NWI) S&T Reports (Dahl, 2005) are the most scientifically defensible sources of national-scale information on wetland location and extent. The NWCA therefore used site-specific information found in the S&T Reports (augmented as detailed below) to identify sampling sites. The sample frame is the FWS National Wetland Status and Trend 2005 survey and was obtained from Tom Dahl at the U.S. FWS. The sample frame consists of all polygons mapped based on 2005 remote sensing information for over 5000 2 mi by 2 mi plots across the 48 states.

Target population

The target population for the NWCA is tidal and nontidal wetlands of the conterminous U.S., including certain farmed wetlands not currently in crop production. The wetlands have rooted vegetation and, when present, open water less than 1 meter deep. A wetland's jurisdictional status[3] under state or federal regulatory programs will not factor into this definition of target.

Working with EPA, FWS created additional plots for the Pacific Coast to help balance the spatial coverage of sites nationally and to ensure the NWCA can produce a representative national assessment of estuarine wetlands. Alaska, Hawaii and the trust territories will not be included in the primary design for the NWCA. Additional attributes added to the sample frame are state, EPA Region, Omernik ecoregion level III, Wadeable Stream Assessment 3 and 9 aggregated ecoregions. The wetland types included are E2EM, E2SS, PEM, PSS, PFO, Pf and PUBPAB which includes PAB, PUB, PUBf, PUBi, PUBn, and PUBu. The following land cover types were excluded: E1UB, E2AB, E2US, LAC, M1, M2, OUT, PUS, RIV, UA, UB, UFP, UO, and URD.

The NWCA design included sites from the following S&T Classes because these classes are very likely to be consistent with the NWCA target population:

- Estuarine Intertidal Emergent
- Estuarine Shrub/Forested
- Palustrine Emergent
- Palustrine Scrub/Shrub
- Palustrine Forested
- Palustrine Unconsolidated Bottom and Aquatic Bed
- Palustrine Farmed

Some wetlands in the S&T Classes listed above will not be consistent with the NWCA target population. These wetlands will most likely be found in the Palustrine Unconsolidated Bottom/Aquatic Bed and Palustrine Farmed classes and will have few, if any, characteristics of naturally-occurring wetlands. If any of these inconsistent sites are selected for sampling, they will be dropped as soon as they are identified (e.g., during desk-top or onsite evaluation).

[3] Impacts to wetlands and other aquatic resources are regulated under the Clean Water Act when an aquatic resource is determined to be a "Water of the United States." Jurisdictional Determinations are made on a case-by-case basis according to the definition found in *40 CFR 230.3(s).* For more information please visit the following website: http://www.epa.gov/owow/wetlands/guidance/CWAwaters.html.

The survey has a two-stage design with the first stage from the FWS National Wetland Status and Trend survey design. It is an area frame design stratified by state and physiographic region where the area frame consists of 2 mi by 2 mi plots that cover the 48 contiguous states. The first stage results in the identification of land cover types focused on wetland types within each 2 mi by 2 mi plot selected (sample size is approximately 5000 plots). The second stage is a Generalized Random Tessellation Stratified (GRTS) survey design for an area resource applied to the stage one sample plots. It is a stratified design with unequal probability of selection based on area within each stratum.

Stratification *is by state* and unequal probability of selection is by seven (7) wetland type categories. Allocation of sites by state and wetland type categories was completed by solving a quadratic programming problem that minimized the sum of the squared deviations of the expected sample size minus proportional allocation of sites by wetland type based on state area within each wetland type subject to constraints that (1) the sum of the expected sample sizes for a state within a wetland type was the following E2EM=128, E2SS=127, PEM=129, PSS=129, PFO=129, Pf=129, and PUBPAB=129, (2) the minimum number of sites for a state was 8, (3) the maximum number of sites within a state for E2EM or E2SS was 13, (4) the maximum number of sites within a state for PEM, PSS, PFO, Pf, or PUBPAB was 10 and (5) the minimum number of sites was greater than or equal to zero for each wetland type and state combination. This approach ensured that the sample size for the seven wetland types was sufficient for national reporting, each state received a minimum number of sites (which also improved the national spatial balance of the sites) and otherwise proportionally allocated the sites by area within a wetland type.

The design includes three panels.

1. Revisit: identifies sites that are to be visited twice.
2. Base: identifies remaining sites to be visited.
3. Over: identifies sites available to be used as replacement sites.

The expected sample size is 900 sites for conterminous 48 states. The maximum number of sites for a state was 69 (Louisiana) and the minimum number of sites for a state was 8 (Vermont). Total number of site visits is 996 allocated to 900 unique sites with 96 sites to be revisited. A 100% over sample size was selected to provide replacement sites that either are not part of the target population or could not be sampled. Sites should be used in SiteID order within each state. If a revisit site cannot be sampled, the next site in the base panel within the state should be used as a revisit site. The map below (Figure 2-1) identifies revisit sites in green, base sites in red and over sample sites in black.

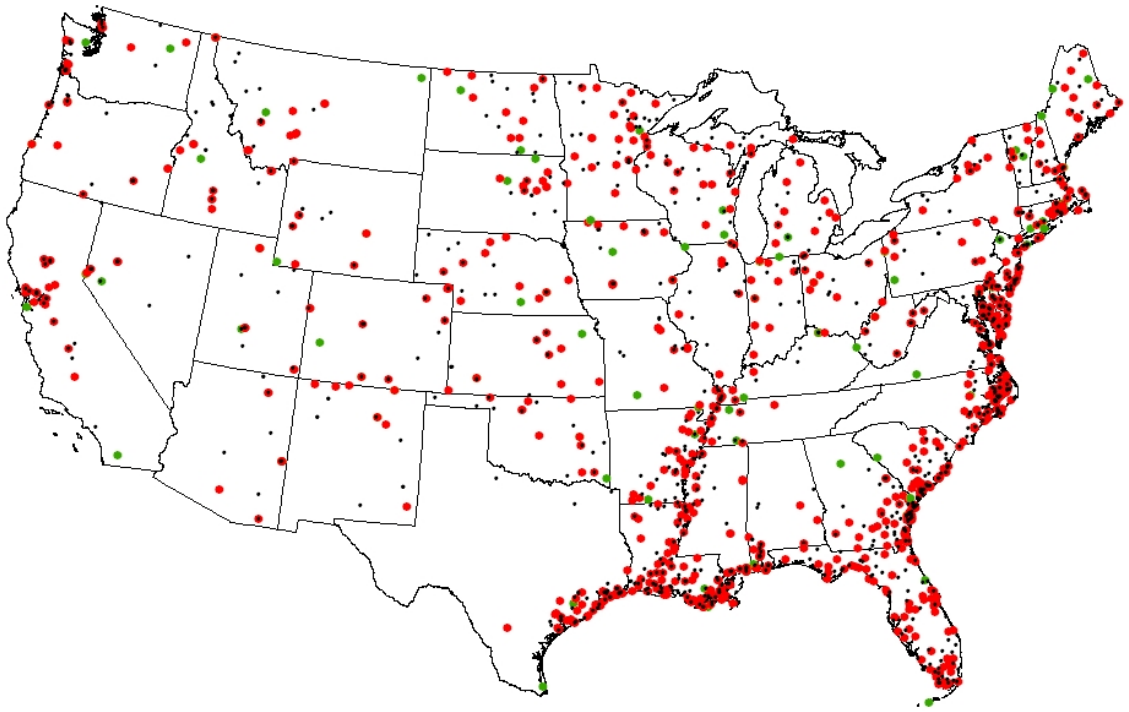


Figure 3-1: NWCA 2011 Survey Design Summary Map

4 INFORMATION MANAGEMENT

Like QA, information management (IM) is integral to all aspects of the NWCA, 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 systems are described in this section.

4.1 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 NWCA 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 NWCA at WED, provide support and guidance to all program operations, in addition to maintaining a central data base management system for the NWCA data.

The central repository for data and associated information collected for use by the NWCA is a secure, access-controlled server located at WED-Corvallis. The general organization of the

information management system is presented in Figure 4-1. 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 NWCA 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 Project Lead. In addition to this inflow, the IM system provides outflow in provision of data files to NWCA staff and other users. The IM staff at WED is responsible for maintaining the security integrity of both the data and the system.

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. Activities to maintain the integrity and assure the quality of the contents of the IM system are also described.

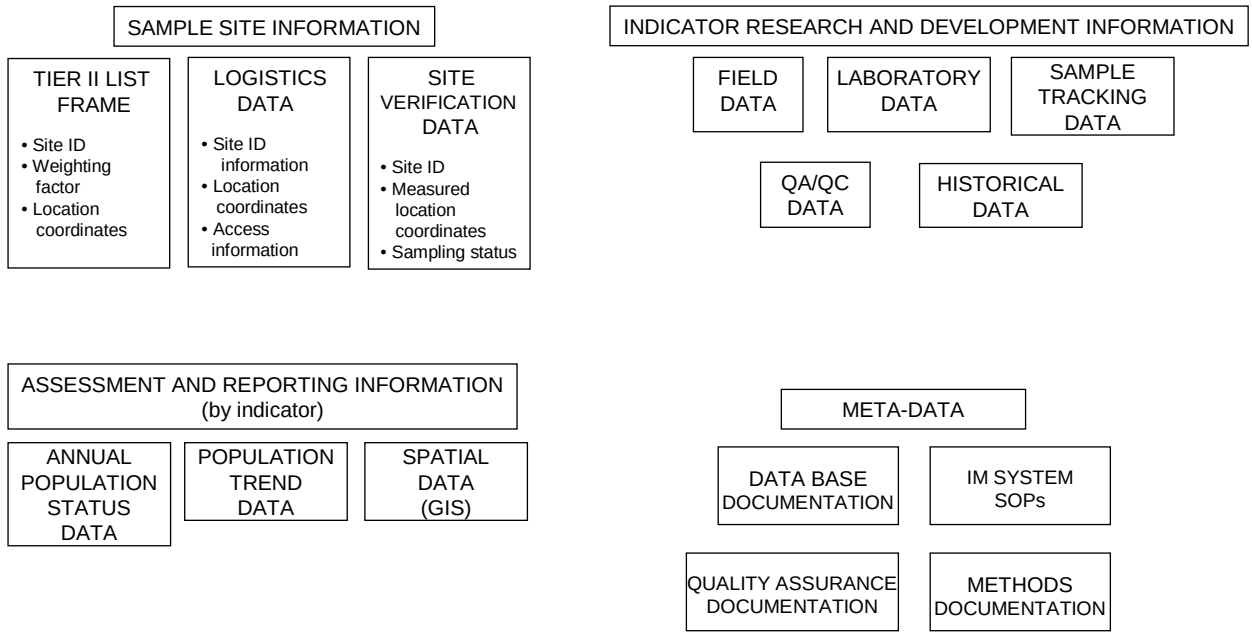


Figure 4-1: Organization of information management system modeled after EMAP Surface Water Information Management (SWIM) system for the NWCA

4.1.1 Design and Site Status Data Files

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, etc.). This “design” data file is provided to the IM staff, Field Implementation

Coordinators, and Field Crew Leaders. Field Crew Leaders determine ownership and contacts for acquiring permission to access each site, and conduct site evaluation and reconnaissance activities. Ownership, site evaluation, and reconnaissance information for each site are compiled into a "site status" data file. Generally, standardized forms are used during reconnaissance activities (see the Site Evaluation Guidelines (USEPA 2011[a]) for more detailed information). 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 Staff for archiving.

4.1.2 Sample Collection and Field Data Recording

Prior to initiation of field activities, the IM staff works with the Project Lead and analytical support laboratories to develop standardized 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.

Data forms are designed in conjunction with IM staff to ensure the format facilitates both field recording and subsequent data entry tasks. All data forms which may be used in the field are printed on water-resistant paper[4]. Copies of the data forms and instructions for completing each form are documented in the Field Operations Manual. Recorded data are reviewed upon completion of data collection and recording activities by a person other than the one who completed the form. The Field Crew Leader checks 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 data forms are transmitted to the IM staff at WED for entry into the central data base management system; the ORD Technical Lead also receives copies of all field-recorded data.

If portable PCs (or handheld data recorders) are to be used in the field, user screens are developed that duplicate the standardized form to facilitate data entry. Specific output formats are available to print data for review and for production of shipping forms. Data may be transferred via modem on a daily basis. Each week CDs containing all down-loaded data for the week are mailed to the Information Management Coordinator (IMC).

All samples are tracked from the point of collection. If field PCs are used, tracking information is entered on custom-designed electronic tracking forms. Hardcopy tracking and custody forms are completed if PCs are not available for use. One copy of the shipping and custody record accompanies all sample transfers; a second copy is transmitted to the IMC and ORD Technical 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 the Field Operations Manual.

[4] Water-resistant paper is not to be copied with photocopiers, as photocopying this type of paper can damage photocopying equipment.

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 4.1-1. Additional QA/QC checks or procedures specific to individual indicators are described in the indicator sections in Section 5 of this QAPP.

Table 4.1-1. 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 on each sample
Data Recording	Data recorded on pre-printed forms of water-resistant paper; field sampling crew reviews data forms for accuracy, completeness, and legibility
Data Qualifiers	Defined qualifier codes used on data form; qualifiers explained in comments section on data form
Sample Custody	Unique sample ID and tracking form information entered in LIMS; sample shipment and receipt confirmed
Sample Tracking	Sample condition inspected upon receipt and noted on tracking form with copies sent to ORD Technical Lead 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 records, including raw data, archived in an organized manner. For example, following verification/validation of the last submission into the NWCA database, it is copied to a terabit external hard drive and sent to the Project Leader for inclusion in his project file, scheduled as 501, permanent records. Processed samples and reference collections of taxonomic specimens submitted for cataloging and curing at an appropriate museum facility

4.1.3 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 Project Lead by telephone.

Most of the laboratory analyses for the NWCA 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 NWCA indicators are described in Table 4.1-2. Additional QA/QC procedures specific to individual indicator analyses are described in the indicator-specific sections 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 NWCA biological samples are described in Table 4.1-3. Additional QA/QC procedures specific to individual parameters are described in the indicator-specific sections of this QAPP.

Table 4.1-2. 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; recalibrate or replace before analyzing any samples
QC Data	Maintain control charts, determine LT-MDLs and achieved data attributes; include QC data summary (narrative and compatible electronic format) in submission package
Data Recording	Use software compatible with NARS-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 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 NARS-SWIM system, documentation of file and data base structures, variable descriptions and formats; summary report of any problems and corrective actions implemented

Table 4.1-3. Biological sample quality control activities

Quality Control Activity	Description and/or Requirements
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 re-identified by different analyst; less than 10% assigned different ID
Taxonomic Reasonableness Checks	Species or genera known to occur in given conditions or geographic area
Reference Collections	Permanent mounts or voucher specimens of all taxa encountered

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 the Field and Laboratory Operations Manuals.

Remaining sample material and voucher specimens may be transferred to EPA's designated laboratory or facilities as directed by the EPA Project Leader. All samples and raw data files (including logbooks, bench sheets, and instrument tracings) are to be retained permanently or until authorized for disposal, in writing, by the EPA Project Leader. (Deliverables from contractors and Cooperators, including raw data, are permanent as per EPA Record Schedule 258. EPA's project records are scheduled 501 and are also permanent.)

4.1.4 Data Review, Verification, and 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 data 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.

The Logistics Coordinator will work with ORD Technical Lead and the IM staff (primary data recipients) to ensure that sufficient QC activities are engaged in the various data management processes. Copies of the raw data files are maintained in the central IM system, generally in active files until completion of reporting and then are transferred to archive files as static data files. Redundant copies are maintained of all data files and all files are periodically backed up.

Some of the typical checks made in the processes of verification and validation are described in Table 4.1-4. 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. They will either be corrected, replaced with a new acceptable value from sample reanalysis, or confirmed as suspect after sample reanalysis. Any suspect data will be flagged for data qualification.

Table 4.1-4. 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 MQOs and project DQOs 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

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. Multivariate techniques have also 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 to validate 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 wetland.

4.2 Data Transfer

Field crews may transmit data electronically via modem or electronic media disc; hardcopies of completed data and sample tracking forms may be transmitted to the IM staff via portable facsimile (FAX) machine or via express courier service. Copies of raw, verified, and validated data files are transferred from the ORD Technical Lead 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 have 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.

4.3 Hardware and Software Control

All automated data processing (ADP) equipment and software purchased for or used in NWCA 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 Technology, Operations, and Planning (OTOP), 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 NWCA 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, NWCA information management staff will assess the impact upon the IM system and develop plans for ensuring timely compliance.

4.4 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 NWCA Cooperators. 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 used. The central data base is backed up and archived according to pre-established procedures. All data records, including raw data, are archived in an organized manner in compliance with EPA and Federal Government records management policies. For example, following verification/validation of the last submission into the NWCA database, all data is copied to a terabit external hard drive and sent to the Project Leader for inclusion in the project file as permanent records. All laboratories generating data and developing data files must have established procedures for backing up and archiving computerized data.

4.5 Data Archive

Ultimately, all data will be transferred to U.S. EPA's agency-wide WQX (Water Quality Exchange) data management system for archival purposes. WQX is a repository for water quality, biological, and physical data and is used by state environmental agencies, EPA and other federal agencies, universities, private citizens, and many others. Revised from STORET, WQX provides a centralized system for storage of physical, chemical, and biological data and associated analytical tools for data analysis. Data from the NWCA project in an Excel format will be run through an Interface Module and uploaded to WQX. Once uploaded, states and tribes will be able to download data (using Oracle software) from their region. In the period after data collection and before transfer to STORET, data will be archived in SWIMS.

5 INDICATORS

As first described in Chapter 2 (Data Quality Objectives, or DQOs), the NWCA has two DQOs: one for condition estimates at the national scale, and the other for the condition estimates within individual ecoregions. The DQO for national-scale estimates is as follows:

> Estimate the proportion of wetlands (± 5%) in the conterminous U.S. that fall below the designated threshold for good conditions for selected measures with 95% confidence.

The DQO for the ecoregions of interest is:

> Estimate the proportion of wetlands (± 15%) in a specific ecoregion that fall below the designated threshold for good conditions for selected measures with 95% confidence.

These two DQOs then govern the structure, performance, archiving, and documentation of all phases of the NWCA.

The influence of these DQOs on the data-acquisition phase (i.e. field sampling) can generally be divided into two types of sub-objectives. The first type of sub-objective influences Field Crew

performance. The second uses repeat visits to evaluate the overall effectiveness of the data collection. The first sub-objective can be stated as follows:

Sub-Objective 1: The Field Crews implement the sampling protocols as designed to collect high quality data.

Sub-objective 1 influences Field Crew operations across all indicators, and applies to both the Crew doing the initial sampling of a POINT and the Crew doing the repeat samples.

QA on the implementation of the protocols by Field Crews would include:

- Appropriate expertise and qualifications (particularly the Botanist);
- Training;
- Site Evaluation and Assistance Visits (i.e. QA audits);
- Review of data forms; and
- Sample tracking.

QA on ability to collect high quality data will focus on completeness (including consideration of proportion of samples that cannot be processed because of problems with how the sample was taken) and accuracy as determined by checks by experts. Two such checks include the check of plant identification on the voucher specimens and the review of soil descriptions by the regional soil scientist.

The second type of sub-objective addresses the overall effectiveness of data collection. This type is summarized in the following two sub-objectives:

Sub-objective 2a: The protocols, when implemented by a Field Crew meeting the QA objective for sampling, generate a reproducible evaluation of ecological condition as demonstrated with data from repeat samples of the same POINT.

Sub-Objective 2b: The data collected by Field Crews meeting the QA objective for sampling can distinguish between sites of different condition, i.e., are robust, in the face of naturally-occurring and sampling variability.

Sub-Objective 2a gets at whether data collected by Field Crews at different times generate the same answers about the condition of the site. This sub-objective influences post-sampling analysis. Examples of QA measures are: (1) The same dominant species are observed in repeat samples ± the MQO and (2) Vegetation structure and composition is not significantly different in repeat samples as determined through the use of multivariate analysis.

Analysis for the Sub-Objective 2b is also done post sampling. It involves developing variance estimates for data metrics which aid in characterizing the utility of metrics through signal to noise ratios, etc. (Stoddard, et al, 2008).

How each of the sub-objectives manifests itself in each of the indicators is detailed in the following sections.

5.1 Vegetation

5.1.1 Introduction

Vegetation is a key attribute of most wetland ecosystems, is sensitive to human-caused disturbance, and accurately reflects wetland condition and biological integrity. It has been used effectively in assessing overall ecological condition and to distinguish particular stressors (Tiner 1999, Garnier et al. 2004, Quétier et al. 2007).

Wetland plant species 1) represent diverse adaptations, ecological tolerances, and life history strategies, and 2) effectively integrate environmental conditions, species interactions, and human-caused disturbance. Data describing species composition and abundance and vegetation structure are powerful, robust, and relatively easy to gather. In addition, they can be used to derive a myriad of metrics or indicators that are useful descriptors of ecological integrity or stress (e.g., USEPA 2002, Bourdaghs et al. 2006, Magee et al. 2008, Mack and Kentula in review). Examples of the types of data to be collected are:

- Species composition and abundance
- Native species
- Alien species
- Floristic quality
- Guild composition
- Community composition
- Vegetation structure

For more detailed information please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

5.1.2 Training and Field Audits

Protocols for collecting data describing species composition and abundance and vegetation structure are provided in the Field Operations Manual (FOM) Vegetation Chapter. Standardized training in implementation of these protocols will be provided to the Botanists and Field Crew members who will assist with botanical data collection to ensure collection of comparable data across the natural study area (see Section 1.3.1.1 for qualifications and duties). In addition, quality assurance audits will be conducted at least once during the field season for each Field Crew to ensure that the protocols are being implemented consistent with training. Ten percent of all sample sites will receive repeat visits to determine if differences exist in field data collection on different days. Revisit sites must be sampled at least 2-4 weeks apart to ensure that we are assessing temporal variability.

5.1.3 Sampling Design

There are two components to collecting vegetation information: the primary component involves field or *in situ* measurement of various species composition indicators; the secondary component involves collecting samples of plant specimens for all unknown species and for 5 vouchers of known species from each site.

The vegetation sampling, observations and associated protocols developed for the NWCA are based on the flexible-plot method of Peet et al. (1998), adapted to meet the objectives and data collection needs of the NWCA. Vegetation sampling will take place in five 100-m^2 plots arranged systematically across the Assessment Area. The FOM Vegetation Chapter includes detailed instructions for establishing the vegetation plots in standard or alternate configurations. Vegetation composition, abundance and structure are assessed at the 100-m^2 scale. Each plot will contain a series of nested quadrats established in two opposing corners to obtain estimates of species diversity, based on species presence at multiple spatial scales (1.0 m^2 and 10m^2).

To optimize vegetation characterization, field sampling for the NWCA will take place during the peak growing season when most vegetation is in flower or fruit. Sampling during this period minimizes seasonal phenological variability and enhances plant species identification accuracy, particularly of difficult species such as grasses and sedges. Although some early ephemeral flowering forbs may be missed by not sampling early in the season, most plant species will be in mature reproductive stages and more readily detected.

On site, it is important to avoid trampling fragile wetland vegetation during sampling activities. Also, to prevent spread of potentially harmful organisms between research sites, all crew members will employ ZERO TAXA TRANSPORT protocols (See FOM Daily Operations Chapter) before leaving the AA. Before entering the vehicle for return to base, field crews are required to decontaminate shoes, clothing and person of all propagules or organisms. Equipment must also be cleaned before replacing it in the vehicle.

5.1.4 Field Measurements and Sampling

5.1.4.1Pre-Sampling Activities

Compiling data forms and organizing the equipment needed for the day's vegetation data collection activities prior to beginning field work enhances efficiency of sampling throughout the rest of the day. Some of this organizational work is completed at the base location or in route to the road location nearest to the POINT.

The vegetation equipment checklist (FOM Vegetation Chapter) ensures all equipment is present. Items should be routinely located in the same places in the vehicle. Keeping the equipment organized by storing and transporting items in the same locations allows items to be easily found, facilitates packing and unpacking the vehicle, minimizes mess and confusion, and helps prevent loss. It is also important to confirm all needed gear and supplies are present before hiking in to the POINT, especially when the location of the POINT is a substantial distance from the nearest road.

Several data forms are used in collecting vegetation data for the NWCA. Each data sheet should be filled out according to the steps outlined in the Vegetation Chapter. Plant specimen labels and plant sample ID tags are also provided. Data forms include:

a. V-1 Vegetation Plot Establishment and Characterization Form
b. V-2 Vascular Species Presence and Cover Form,
c. V-3 Ground Surface Attributes Form,
d. V-4 Snag and Tree Counts and Tree Cover Form,

5.1.4.2Sampling Activities

All field measurement and sampling operations will be conducted by a Vegetation (Veg) Team consisting of a Botanist/Ecologist and Botanist Assistant.

All measurements and observations are recorded on standardized forms which are later entered in to the central National Aquatic Resource Surveys (NARS) surface waters information management system at WED-Corvallis. Table 5.1-1 provides a brief summary of the observations recorded by the Veg Team.

Table 5.1-1. Field measurement methods: vegetation

Variable or Measurement	Units		Summary of Method	
Vascular strata coverage	%		Estimate total cover of emergent and non-aquatic vegetation by height class (< 0.5, 0.5-2, 2-5, 5-15, 15-30, >30 m, or liana, vines, and epiphytes), submerged aquatic vegetation and floating aquatic vegetation	
Non-vascular coverage	%		Estimate percent cover for non-vascular taxonomic groups (bryophytes, ground lichens, arboreal epiphytic bryophytes and lichens, filamentous or mat-forming algae,and macro algae)	
Individual vascular coverage	%		Estimate percent cover for each species and record the predominant height class in which it occurs	
Ground surface attributes: coverage	%		Estimate cover of water, bareground, vegetative litter, and dead woody debris	
Ground surface attributes: depth	cm		Measurements for water (minimum, predominant, and maximum depth) and vegetative litter	
Tree coverage	%		Estimate percent cover for each species by height class (< 0.5, 0.5-2, 2-5, 5-15, 15-30, >30 m)	
Tree count	None		Count stems for individuals >5 cm diameter breast height (dbh) by diameter class (5-10, 11-25, 26-50, 51-75, 76-100, 101-200 and > 200 cm), by species	
Standing dead trees and snags	None		Count total number of stems >5 cm dbh by diameter class (5-10, 11-25, 26-50, 51-75, 76-100, 101-200 and > 200 cm), by species	
Species presence data	None		For each species present, record the smallest quadrat in which it occurs	

General Cover Estimation Protocols:
The entire range of values from 0 to 100% may be used when estimating cover for a species or other entity within each 100-m^2 plot. However it is not necessary or appropriate to deliberate extensively over small differences in values for cover estimates (e.g., 0.1% or 0.5%, 5 or 7%, 25 or 30%, 75% or 85%). This degree of precision is likely to exceed the accuracy of the Botanist/Ecologist's ability to detect cover differences over the area of the module. See Table 5.1-2 for guidelines on increments of resolution for different cover ranges.

Table 5.1-2. Guidelines for resolution when estimating percent cover

For Cover Of:	Use % Increments Of:
Trace (<1%) = 0.1%	NA
1 to 5%	1%
5 to 25%	5%
25 to 50%	5 to 10%
55 to 100%	10 to 15%

Nomenclature:
To effectively identify plant species in the field and to key unknown taxa, it will be necessary to use local floras appropriate to each region or state. This means numerous taxonomies will be applied across the 48 conterminous states comprising the study area. To reduce potential discrepancies in nomenclature, it is suggested that each Botanist/Ecologist reconciles species names to the USDA PLANTS nomenclature.

Collecting Plant Material for Specimens:
Throughout the sampling day, the Botanist/Ecologist and Botanist Assistant collect all unknown plant species and five known plant species (randomly selected from species identified in the 100-m^2 vegetation plots) from the site. Specimens are carefully labeled with tracking information and placed in a plant press to dry. The Veg Team ensures that all tracking information always remains with the specimens (pertinent information written on the Plant Specimen Tag and affixed on the newspaper sleeve containing the specimen and on the Plant Specimen Label Form). Detailed instructions for specimen collection, pressing, labeling, shipping, and tracking are found in the FOM Vegetation Chapter. Voucher specimens should not be collected for plants that are rare within the vegetation plot or Assessment Area.

Pressing Plant Specimens:
Plant specimens represent *critical* vegetation data; thus, it is important to press plant material as soon after collection as practicable to preserve the morphological features of the plant for later identification by a botanist. In those situations where important morphological features may be damaged or lost by pressing and drying (i.e., flower color, fruit color, and fruit shape) it is important to document these features on the plant specimen label form. Plant specimen labels are considered field data, and a plant specimen is incomplete in the absence of accurate label data. A completed **Plant Specimen Label Form** should be enclosed in the newsprint folder of each specimen. Photographic documentation is also valuable. The field day is not considered finished until all plant specimens collected in the field are properly pressed and labeled. Key elements of label data and steps for pressing plant specimens can be found in the appropriate sections of the FOM Vegetation Chapter.

Drying, Storing, and Shipping Plant Specimens:
Normally, pressed plant specimens should be thoroughly dried before removing them from the presses. Ideally, full plant presses will be returned to the base location after a few field days and placed on plant dryers to dry. Once the specimens are dry they can be removed from the press and shipped to an expert for identification or stored for later identification by the Veg Team during non-field days of the field season.

For crews that work for more than four or five days in the field without returning to a location where plants can be dried, wet plant specimens may need to be removed from the presses, packaged and shipped to a location where the specimens can be dried and processed.

The steps for handling specimens once they are in the press can be found in the appropriate section of the FOM Vegetation Chapter.

5.1.4.3Quality Assurance Objectives

As mentioned above in section 2.2.2 (Precision, Bias, and Accuracy), precision of field measurements will not be monitored during the NWCA. Previous plant identification experience or class work will be valuable for Veg Team members, but mandatory NWCA training will prepare the crew to accurately complete vegetation data collection tasks according to the standardized field protocols.

MQOs are given in Table 5.1-4. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.1-4 represent the maximum allowable criteria for statistical control purposes. Precision is determined from results of revisits (field measurements) taken on a different day (at least 2-4 weeks apart).

5.1.5 Laboratory Methods

For the purposes of this manual a **Herbarium** represents the person identifying and processing unknown specimens collected in the field. This could be a field botanist, state identified herbarium, EPA identified regional herbarium, or National EPA Contractor. The Herbarium is responsible for ensuring all plant identification and processing tasks outlined in this manual are completed. In some cases this may require the Herbarium to identify partners to assist with the work. The Herbarium identifies all unknown plant species.

Known plant species collected for quality assurance are sent to the **QA Herbarium** for re-identification. A QA Herbarium is an independent qualified botanist, state or EPA identified regional herbarium that agrees to use the NWCA prescribed methods to ensure that all QA vouchers receive the same level of taxonomic precision. Ten percent of unknown specimens identified by the Herbarium are also sent to the QA Herbarium for re-identification and quality assurance. Details on how the Herbarium and QA Herbarium should handle and identify plant specimens can be found in appropriate section of the LOM Vegetation Chapter. Voucher specimens will arrive at the QA Herbarium without a species name. The QA Herbarium will then blindly re-identify all species to ensure that the identifications are independent.

Voucher and unknown specimens may arrive at the Herbarium or QA Herbarium either dried and pressed, or pressed and possibly still wet in the plant press. If specimens are pressed and dried they must be treated for contamination (detritivores, molds, and pests) before identification. If specimens are still wet in the plant press they must be dried and treated for

contamination before identification (LOM Vegetation Chapter). It will be important to maintain a record of specimen custody through shipping to identification to data entry to ensure the correct species name is recorded for the appropriate NWCA site and Vegetation Plot in the database.

Tracking Specimens:

In the field, each voucher specimen collected is assigned a set of tracking information, which is recorded on the Plant Specimen Tracking Form. If a specimen does not have any of the necessary information, contact the Logistics Coordinator as soon as possible. It is important that every specimen sent to and received by the lab is tracked following the protocols described in the appropriate section of the LOM Vegetation Chapter. Specimens may follow one of the paths described in Figure 5-1.

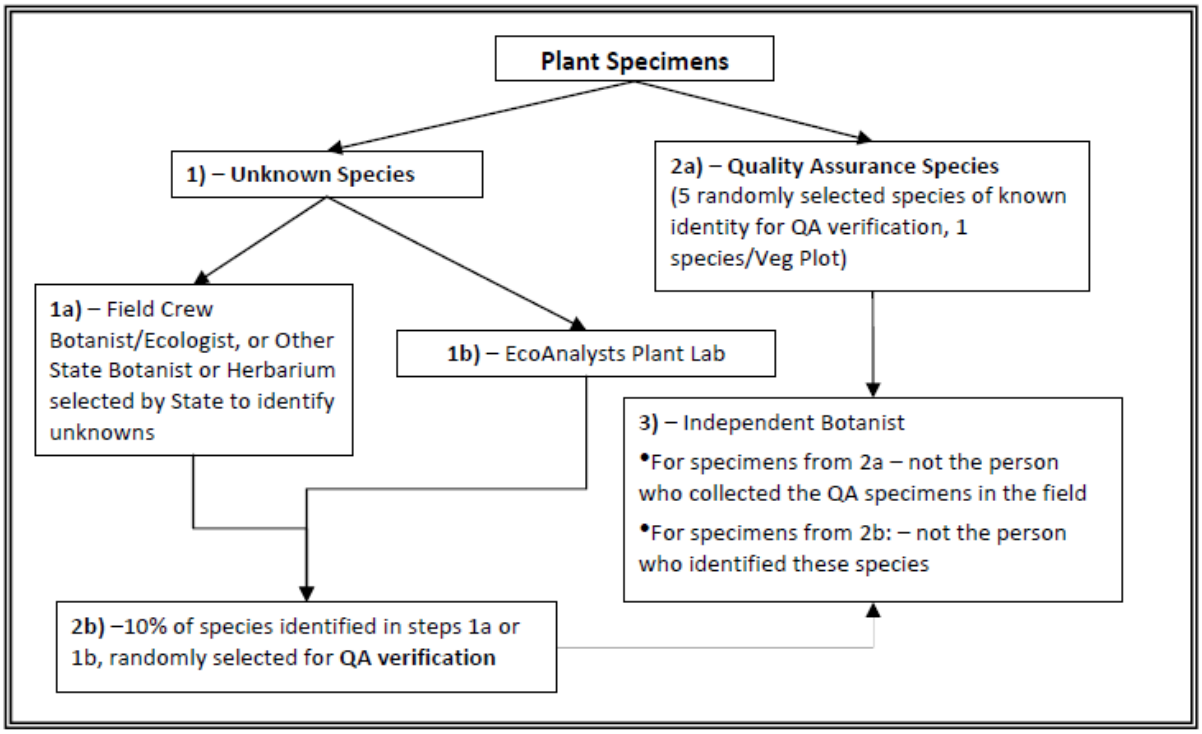


Figure 5-1: Potential options for plant vouchers collected as part of the 2011 NWCA

Nomenclature:

This means numerous taxonomies will be applied during the 2011 NWCA. The Herbarium and QA Herbarium will reconcile all species received to the standard found in USDA Plants. The LOM Vegetation Chapter contains more information on reconciling taxonomy to USDA Plants.

5.1.6 Quality Assurance Objectives

MQOs are given in Table 5.1-3. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.1-4 represent the maximum allowable criteria for statistical control purposes. Precision is determined from results of revisits (field measurements) taken on a different day (at least 2-4 weeks apart).

Table 5.1-3. Measurement data quality objectives: vegetation indicator

Variable or Measurement	Precision	Taxonomic Disagreement	Completeness
Field Measurements and Observations	±10%	≤ 15%	90%
NA = not applicable in most cases. This would apply if the field auditor did a separate assessment and compared the results to the crews.			

5.1.7 Quality Control Procedures: Field Operations

Precision, bias and accuracy of field measurements will not be monitored during the NWCA[5]. Control measures to minimize measurement error among crews and sites include the use of standardized field protocols, consistent training of all crews, field assistance visits to all crews, and availability of experienced technical personnel during the field season to respond to site-specific questions from field crews as they arise.

Upon completion of sampling, the Botanist/Ecologist reviews all vegetation forms for completeness, legibility, and for any errors in species names.

The Botanist/Ecologist checks the voucher collection record on the Vascular Plant Species Presence and Cover Form (FOM Vegetation Chapter) for all taxa with pseudonyms to ensure that specimens have been collected for all unknown species. Additionally, the Botanist/Ecologist and Botanist Assistant collect 5 known plant species (randomly selected from species identified from the 100-m^2 vegetation plots) as voucher specimens. These voucher specimens will be sent to a QC taxonomist for re-identification.

1. The QC taxonomist will perform re-identifications completing a copy of the Vegetation Taxonomic Bench Sheet for each specimen. Each bench sheet must be labeled with the term “QC ID.” As each bench sheet is completed, it must be faxed or emailed to the project facilitator.

[5] Bias, for example, cannot be determined directly, since the “true” values at any particular site are not known.

2. The project facilitator will compare the taxonomic results generated by the primary and QC taxonomists for each specimen and calculate percent taxonomic disagreement (PTD) as measures of taxonomic precision (Stribling et al. 2003) as follows:

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

Equation 1

where

$comp_{pos}$ = the number of agreements (positive comparisons)
N = the total number of specimens in the larger of the two counts.

3. Unless otherwise specified by project goals and objectives, the measurement quality objective for enumerations will be a mean PTD less than or equal to 15, calculated from all the specimens sent to the QC taxonomist. Results greater than these values will be investigated and logged for indication of error patterns or trends, but all values will generally be considered acceptable for further analysis, unless the investigation reveals significant problems.

4. Corrective action will include determining problem areas (taxa) and consistent disagreements, addressing problems through taxonomist interactions. Disagreements resulting from identification to a specific taxonomic level, creating the possibility to double-count "unique" or "distinct" taxa will also be rectified through corrective actions.

5. The project facilitator will prepare a report or technical memorandum. This document will quantify both aspects of taxonomic precision, assess data acceptability, highlight taxonomic problem areas, and provide recommendations for improving precision. This report will be submitted to the project manager, with copies sent to the primary and QC taxonomists and another copy maintained in the project file.

Ten percent of all sites will receive repeat sampling visits to be sampled by a Field Crew to determine the extent to which the population estimates might vary if they were sampled at a different time (revisit sites must be sampled at least 2-4 weeks apart).

5.1.8 Quality Control Procedures: Laboratory Operations

A subset of plant samples collected as unknowns and later identified by the lab will need to be verified by a QA taxonomist for additional Quality Assurance. The lab will randomly select 10% of the identified unknown samples to be sent to the QA taxonomist, another experienced taxonomist who did not participate in the original identifications. A chain-of-custody form will be completed and sent with the specimens.

6. The QC taxonomist will perform re-identifications completing another copy of the Vegetation Taxonomic Bench Sheet for each specimen. Each bench sheet must be labeled with the term "QC Re-ID." As each bench sheet is completed, it must be faxed to the project facilitator.

7. The project facilitator will compare the taxonomic results generated by the primary and QC taxonomists for each specimen and calculate percent taxonomic disagreement (PTD) as measures of taxonomic precision (Stribling et al. 2003) as follows:

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

Equation 1

where

$comp_{pos}$ = the number of agreements (positive comparisons)
N = the total number of specimens in the larger of the two counts.

8. Unless otherwise specified by project goals and objectives, the measurement quality objective for enumerations will be a mean PTD less than or equal to 15, calculated from all the specimens in the 10% set sent to the QC taxonomist. Results greater than these values will be investigated and logged for indication of error patterns or trends, but all values will generally be considered acceptable for further analysis, unless the investigation reveals significant problems.

9. Corrective action will include determining problem areas (taxa) and consistent disagreements, addressing problems through taxonomist interactions. Disagreements resulting from identification to a specific taxonomic level, creating the possibility to double-count "unique" or "distinct" taxa will also be rectified through corrective actions.

10. The project facilitator will prepare a report or technical memorandum. This document will quantify both aspects of taxonomic precision, assess data acceptability, highlight taxonomic problem areas, and provide recommendations for improving precision. This report will be submitted to the project manager, with copies sent to the primary and QC taxonomists and another copy maintained in the project file.

5.1.9 Data Management, Review, and Validation

The Botanist and Field Crew Leader are responsible for the validity of all field-generated data (i.e. measurement and observation data) up to the point it is sent to EPA (ORD/Corvallis). The Botanist and Field Crew Leader are likewise responsible for the proper labeling, storage, and delivery for shipping of all voucher samples, and for informing ORD/Corvallis when samples have been shipped. Laboratory SOPs (see Chapter 2 for details) will be followed to ensure that data generated and delivered to EPA are valid. Once data have been delivered to EPA, data quality (DQ) procedures (as detailed in Chapter 2) will be followed to ensure the validity of data in storage, analysis, reporting and archiving. All raw data (including all standardized forms and logbooks) are retained permanently in an organized fashion in accordance with EPA records management policies.

5.2 Soils

5.2.1 Introduction

The presence of hydric soil is a defining characteristic of wetland ecosystems. Soils influence surface and ground water movement in wetlands. Soils also provide a matrix for biogeochemical

processes (e.g., nutrient cycling, pollutant storage) which affect wetland vegetation and other wetland ecosystem components that reflect ecological condition (Tiner 1991, Mitsch and Gosselink 2007). Examples of the types of data to be collected are:

- Hydric soil field indicators
- Description of site, soil morphology, and other characteristics
- Soil chemistry
- Soil isotope and sediment enzymes
- Bulk density

For more detailed information please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

5.2.2 Sampling Design

The soil sampling, observations and associated protocols used in the NWCA were chosen in consultation with USDA soil scientists, as well as wetlands scientists and field sampling experts in the EPA, states, tribes, academia and the private sector. For the soil indicator class, individual metrics were chosen to:

- Describe current physical and chemical properties of the soil;
- Identify the presence of hydric soils (which inform soil development and history); and
- Ascertain the presence and extent of disturbance which affects soil function.

There are two components to collecting soil information: The first component involves field measurement and description of soil macromorphological properties, e.g., texture, color, and structural attributes; the second component involves collecting soil samples for laboratory analysis of various physical characteristics and chemical constituents (NWCA FOM, 2009).

As described in Section 1.3.1 (Overview of Field Operations) above, NWCA Field Crews will be divided into two teams, the Veg Team (2 members) and the AB Team (2 members) (NWCA FOM, 2009). The AB Team will be responsible for collecting Soil Indicator samples and site descriptions.

After the Veg Team has delineated the Vegetation plots (See Section 5.1 above), soil-related sampling will be conducted at four soil pits, located at the southeast corner of the vegetation plot. The Soils Chapter of the FOM details how the four pits will be located, as well as detailed procedures for completing the protocols; equipment and supplies required are also listed. Two activities will take place 1) description of the soil and 2) collection of soil samples for laboratory analysis.

Soil profile information will be the first data collected at the pit, from a 25cm x 10cm x 60cm slab. Soil samples will be collected at one pit, chosen as representative of the soil in the AA, only after soil profile information is recorded. As detailed in the FOM Soils Chapter 6, there are 3 distinct soil sample collection protocols:

1. Samples collected for physical, chemical and nutrient analysis
2. Samples collected for soil isotope and sediment enzyme analysis
3. Samples collected for determination of bulk density (Db)

A modified 60 mL plastic syringe will be used to collect three isotope samples and six enzyme core samples from three locations around the unexpanded representative pit. The isotope sample will be placed in a clean quart-size zip lock plastic bag and the enzyme sample will be placed in a gallon-sized zip lock bag. Each of the bags is pre-labeled (using an indelible pen) with the Site ID #, date, and sample number.

Bulk density and chemistry samples are collected from each soil horizon that is greater than 8 cm thick. A special bulk density sampler is used to collect three cores of known volume of sample for bulk density, that are placed, together, in a pre-labeled soil bag. The label on the outside of the bag will contain the depth of the horizon. Another label is stapled on the outside of the bag that contains the diameter and length of cores and volume of sample.

Chemistry samples are collected simultaneously with bulk density samples. When the corer is placed, loose soil from the same horizon is broken off and placed in the pre-labeled soil bag. The label will contain the depth of the horizon. If large rocks are removed from the sample, the estimated percent volume that they made up should be recorded on the second label that is stapled to the outside of the bag.

Tools will be wiped clean between sampling pits to prevent contamination of soils collected at different horizons, soil pits, and sites.

On site, it is important to avoid trampling fragile wetland vegetation during soil sampling activities. Also, to prevent spread of potentially harmful organisms between research sites, all crew members will employ ZERO TAXA TRANSPORT protocols (See FOM Daily Operations Chapter) before leaving the AA, and before entering the vehicle for return to base. Decontaminate shoes, clothing and person of all propagules or organisms. Clean equipment before replacing it in vehicle.

Shipping protocols differ for the different soil samples. Soil enzyme and isotope samples are shipped on ice within 24 hours of collection. Bulk density and chemistry samples should be kept cool and can be held for up to two weeks and batched for shipping..

Shipping and receiving regulated soils. Soils that may contain pests (i.e., bacteria, plant viruses, fungi, nematodes, and life stages of destructive mollusks, acari, and insects) are regulated by U.S Department of Agriculture's Animal and Plant Health Inspection Service (APHIS). Areas within states that are under Federal quarantine must follow the conditions and safeguards prescribed by APHIS before shipping to another part of the country. To ensure that the national survey is in compliance with APHIS recommendations all soils collected for the survey will be shipped as regulated soils. Participating labs are responsible for obtaining and maintaining a valid permit for receiving regulated soils (USDA APHIS PPQ 525-A, Figure 5-2 below).

Upon arrival at the lab, soil samples will be separated into regulated and non-regulated based on their county and state of origin (as recorded on the water proof label affixed to the outside of

the sample bag). The lab is responsible for following all APHIS protocols when handling or disposing of regulated soils found in *7 CFR 330*.

APHIS Animal and Plant Health Inspection Service

Plant Protection & Quarantine

United States Department of Agriculture
Animal and Plant Health Inspection Service
4700 River Road
Riverdale, MD 20737

Permit to Receive Soil

Regulated by 7 CFR 330

This permit was generated electronically via the ePermits system.

PERMITTEE NAME:	Dr. Thomas Reinsch	**PERMIT NUMBER:**	P330-08-00009
COMPANY:	USDA-NRCS-NSSC	**APPLICATION NUMBER:**	P525-071002-007
RECEIVING ADDRESS:	Federal Building, Room 152, MS 41 100 Centennial Mall North Lincoln, NE 68508-3866	**DATE ISSUED:**	01/14/2008
MAILING ADDRESS:	Federal Building, Room 152, MS 41 100 Centennial Mall North Lincoln, NE 68508-3866		
PHONE:	(402) 437-4179		
FAX:	(402) 437-5760	**EXPIRES:**	**01/14/2011**

PORTS OF ARRIVAL/PLANT INSPECTION STATIONS: Various Ports of Entry Staffed by CBP-Agriculture Inspection

HAND CARRY: No

Under the conditions specified, this permit authorizes the following:

Quantity of Soil per Shipment and Treatment
Over 3 lbs

PERMIT CONDITIONS

1. This permit authorizes the importation of soil, under the conditions specified below. Upon arrival in the United States, the articles, shipping container(s), and paperwork are subject to inspection by officials of Customs and Border Protection, Agriculture Inspection (CBP-AI) and the USDA, Plant Protection and Quarantine (PPQ).

2. Under the Plant Protection Act, individuals or corporations who fail to comply with the following conditions and authorizations, or who forge, counterfeit, or deface permits or shipping labels will receive civil or criminal penalties, and will have all current permits cancelled and future permit applications denied.

3. Any person who unloads, lands, or otherwise brings or moves into or through the United States any regulated plants, plant products, plant pests, soil or other products or articles in violation of the regulations will be subject to prosecution under the applicable provisions of the law.

Permit Number P330-08-00009

THIS PERMIT HAS BEEN APPROVED ELECTRONICALLY BY THE FOLLOWING PPQ HEADQUARTER OFFICIAL VIA EPERMITS.	DATE
Maria Corpuz	01/14/2008

WARNING: Any alteration, forgery or unauthorized use of this Federal Form is subject to civil penalties of up to $250,000 (7 U.S.C.s 7734(b)) or punishable by a fine of not more than $10,000, or imprisonment of not more than 5 years, or both (18 U.S.C.s 1001)

APHIS Animal and Plant Health Inspection Service

Plant Protection & Quarantine

4. All foreign cargo of agricultural interest is inspected at the first port of arrival or the first port of unlading. If a shipment arrives at a port without the required official personnel available to do the proper inspection, and/or treatment, any subsequent movement, or any transfer and/or transloading, must be approved by CBP-AI.

5. A copy of this permit must accompany all shipments authorized under this permit.

6. The soil is to be shipped in sturdy, leak-proof containers.

7. CBP-AI and PPQ have the option to order and approve treatment, re-exportation or destruction of a shipment, a portion of a shipment, or any other material associated with the shipment (i.e. pallets, packaging, means of conveyance). This will be done if the official personnel find that the shipment requires treatment as a condition of entry, is contaminated with a quarantine plant pest or pests, is commingled with prohibited plant material, or if required documentation is incomplete or missing.

8. The shipment must be free from foreign matter or debris, plants, noxious weed seeds, and living organisms such as parasitic plants, pathogens, insects, snails, and mites. Material found to be commingled with unauthorized material will be subject to the same action (i.e. re-export, destruction) as the unauthorized material.

9. All solid wood packing material (SWPM) present with this shipment must be in compliance with ISPM 15 treatment and IPPC stamp requirements and enforcement. Noncompliant shipments will be treated, re-exported or destroyed at the consignee's expense.

10. All costs and arrangements for the safeguarding of the cargo and the transportation of the cargo are the responsibility of the importer, broker, or other parties associated with the shipment.

11. The shipment can be released without treatment at the port of entry to the permittee's address listed on the permit or label, or an authorized user only if the final destination is an approved facility listed at https://web01.aphis.usda.gov/PPQ/AuthSoilLabs.nsf/web?openform.

12. Permit is to be utilized by the permittee or authorized user only (authorized users must present a written, dated, and signed statement on letterhead from the permittee, along with a valid ID and a copy of this permit).

13. There is no further distribution of soil without prior approval from the State and Federal Regulatory Officials. Soil is to be used strictly for analysis in a laboratory environment at USDA-NRCS-NSSC located in Lincoln, NE.

14. Upon receipt, all samples will remain within the approved soil laboratory identified on this permit. Laboratory access is restricted to individuals authorized by the permit holder.

15. This permit does not authorize the use of soil for growing purposes and or the isolation or culture of organisms sourced from imported soil.

16. All unconsumed soil, containers, and effluent is to be autoclaved, incinerated, or properly sterilized by the permittee at the conclusion of the project as approved and prescribed by PPQ in the compliance agreement.

17. Valid for shipments of soil not heat treated at the port of entry, only if a Compliance Agreement (PPQ Form 519) has been completed and signed. Compliance Agreements and Soil Permits are non-transferable. Notify local USDA office promptly if the permittee leaves the company.

18. This permit authorizes shipments from all foreign sources, including Guam, Hawaii, Puerto Rico, and the U.S. Virgin Islands through any U.S. port of entry.

END OF PERMIT CONDITIONS

Permit Number P330-08-00009

THIS PERMIT HAS BEEN APPROVED ELECTRONICALLY BY THE FOLLOWING PPQ HEADQUARTER OFFICIAL VIA EPERMITS.	DATE
Maria Corpuz	01/14/2008

WARNING: Any alteration, forgery or unauthorized use of this Federal Form is subject to civil penalties of up to $250,000 (7 U.S.C.s 7734(b)) or punishable by a fine of not more than $10,000, or imprisonment of not more than 5 years, or both (18 U.S.C.s 1001)

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Figure 5-2: Example PPQ 525-A Regulated Soils Permit

5.2.3 Sampling and Analytical Methods

Field Observations and Sampling:

Field measurement and sampling operations will be conducted by the AB Team. Field measurements collected are described in Table 5.2-1 below. All observations are recorded on standardized forms which are later entered in to the central NARS surface waters information management system at WED-Corvallis.

Table 5.2-1. Field measurement methods: soil profile metrics.

Variable or Measurement	Units		Summary of Method	
For each pit				
Date	NA		Date of observations	
Location	d, m, s		Latitude & longitude, from GPS	
depth	cm		Depth of profile observations	
Hydric soil indicators	NA		Compare the soil texture as determined for the soil profile horizons (see below) to the descriptive keys in both generic and regionally specific versions of the Field Indicators of Hydric Soils in the United States (USDA and NRCS 2006).	
Water level	cm		Depth to water from soil surface	
Saturation level	cm			
For each of the top 7 (O, A, E, B, C, L, R) soil horizons				
Horizon depth	cm		Measure the depth of each soil horizon	
Texture	NA		Simple hand test described in FOM Figure 9-4 (NRCS, 2009. Modified from S.J. Thien. 1979)	
Matrix color	NA		Compare moist color to color-chips from the Munsell Color Book	
Redoximorphic features	NA			
Concentrations or deletions color	NA		Same as matrix color method	

As mentioned in Section 5.2-2, soil samples will be collected for soil istopes, enzymes chemical and nutrient analysis, and determination of Db. For bulk density and chemistry, separate samples will be collected for each soil horizon measuring greater than 8 cm from the representative pit, down to 1.25m, if possible. Confirmation of sample collection status will be recorded on the standardized form.

Table 5.2-2. Soil Sample Collection

Sample Type	Summary of Method	
Soil isotope	One core each from each of 3 locations, from the uppermost horizon, around unexpanded soil pit	
Soil enzyme	Two cores from each of three locations, from the uppermost horizon, around unexpanded soil pit	
Bulk density	Three cores of known volume from each soil horizon that measures more than 8 cm, to 125 cm	
Chemistry	Approximately 1 to 1.5 L of sediment from each soil horizon that measures more than 8 cm, to 125 cm	

All receipts and records of shipping will be kept as part of the permanent record of the NWCA and copies of the pertinent **NWCA Soil Sample Form(s)** will be included with shipped samples.

5.2.4 Quality Assurance Objectives

As mentioned above in section 2.2.2 (Precision, Bias, and Accuracy), precision of field measurements will not be monitored during the NWCA. Previous soils experience or class work will be valuable for AB team members, but mandatory NWCA training will provide an understanding of basic soil processes, soil description methods, and sampling techniques. This training will prepare the crew to accurately complete soil data collection tasks according to the standardized field protocols.

MQOs are given in Table 5.2-2. 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. Precision is determined by the comparison of field measurements from two visits to the same site; the revisit is at least 2-4 weeks after the first visit. Due to the high level of disturbance caused by the soil sampling methods, it is not appropriate for the soil protocols to be completed in the same location twice. During the second sampling event the AB team will locate the soil pits as close to the original soil pit locations as possible without entering into the zone of disturbance created by the first sampling event. This will ensure that the soil data collected are as similar to the original data as possible.

Table 5.2-3. Measurement quality objectives: soil indicator

Variable or Measurement	Precision	Accuracy	Completeness
Field Measurements and Observations	±10%	NA	90%
NA = not applicable in most cases. This would apply if the field auditor did a separate assessment and compared the results to the crews.			

5.2.5 Quality Control Procedures: Field Operations

Control measures to minimize measurement error among crews and sites include the use of standardized field protocols, consistent training of all crews, and availability of experienced technical personnel during the field season to respond to site-specific questions from field crews. Additionally, field crews will apply a consistent labeling convention across all samples (see FOM Soils Chapter for details on info to include on labels).

Other controls include audits and revists. Quality assurance audits are conducted of each Field Crew at least once during the field season, to ensure the protocols followed are consistent with training. Ten percent of all sites will receive repeat sampling visits to be sampled by a Field Crew to determine the extent to which the population estimates might vary if they were sampled at a different time.

In addition, field Crew Leaders are responsible for reviewing all forms for completeness and legibility, and ensuring that all samples are properly collected and shipped. Field forms are then sent to participating NRCS State Soil Scientists to ensure that horizon designations are correct. Specific quality control measures are listed in Table 5.2-3 for field measurements and observations.

Table 5.2-4. Field quality control: Soil indicator

Quality Control Activity	Frequency	Acceptance criteria	Corrective Action
Quality Control			
Check completeness of soil descriptive data	Each soil horizon	Values for each soil horizon	Repeat observations
Check for completeness of soil sample collection for chemical analyses and bulk density	Each station	Data sheets complete where appropriate	Repeat observations
Sample Storage	Each station	Nontidal soils: samples kept in a cool dry place until shipped Tidal soils: samples kept on ice until placed in a refrigerator or shipped with cold packs	Qualify sample as suspect for all analyses
Data Validation			
Estimate precision of measurement based on repeat visits	2 visits	Measurements should be within 10 percent	Review data for reasonableness; Determine if acceptance criteria need to be modified

5.2.6 Quality Control Procedures: Laboratory Operations

Standardized lab protocols, consistent training of all lab technicians, lab assistance visits to all labs, and availability of experienced technical personnel to respond to site-specific questions as they arise are important to ensuring the quality of lab data. Additionally, control measures to minimize measurement error among lab technicians and laboratories include the use of a Control Sample, a Blank Sample, Data Review, and Data Validation.

A **Control Sample** represents a sample of known concentration for a particular attribute. A Control Sample is collected in bulk for an attribute and repetitively analyzed to determine statistical control limits (i.e., range of expected values) for the particular method. A Control Sample is analyzed in conjunction with every batch of samples to ensure the method was run correctly. If the value of the Control Sample falls outside the expected range of values then the process has failed and the batch is triggered for reanalysis.

A **Blank Sample** is used to ensure equipment is thoroughly cleaned before each use. A Blank Sample is especially important when measuring soil chemistry (i.e., trace metals) because concentrations may be quite small. A Blank Sample is analyzed in conjunction with every batch of samples to ensure that proper equipment cleaning protocols are followed. If the value of the Blank Sample does not equal zero or fall below the MDL, then the equipment is not clean and the batch is triggered for reanalysis.

The process of **Data Validation** is described here. Laboratory data undergo four **Data Reviews**, first by the Bench Analysts, second by the Lead Analyst, third by the Project Coordinator Soil Scientist, and fourth by a Soil Scientist Liaison with expertise in soils from the region where the samples are from. The Bench Analysts verifies that blank and control samples return results that fall within established control limits. The Lead Analyst examines the data for inconsistencies and apparent anomalies; inconsistencies usually take the form of unexpected high or low values for a particular analyte or values that do not fit with the expected trend of a soil profile. The Project Coordinator will use professional judgment to determine whether the project data are self-consistent and congruent with the site data collected in the field; incongruities within the data that can be explained either by site data or the results of other analytes are recorded. A final review is given by a Soil Scientist Liaison to the area of sample origin, before the data are released.

Table 5.2-5. Lab analysis quality control: soils indicator

Activity or Procedure	Requirements and Corrective Action
Range check of Control Sample	If value is outside expected range, batch is triggered for reanalysis
Value check of Blank Sample	If value is >0 or the MDL, batch sample is triggered for reanalysis
Data Review	Corrective reporting for explicable incongruities within the data
Data Validation	Corrective reporting for explicable incongruities within the data

5.2.7 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.2-5. The Field Crew Leader is responsible for the validity of all field-generated data (i.e. measurement and observation data) up to the point it is sent to EPA (ORD/Corvallis). The Field Crew Leader is responsible for the proper labeling, storage, and delivery for shipping of all samples. The Field Crew Leader is responsible for notifying both the laboratory and ORD/Corvallis when samples have been shipped. Laboratory SOPs (see Chapter 2 for details) will be followed to ensure that data generated and delivered to EPA are valid. Once ORD/Corvallis receives the data, DQ procedures (as detailed in Chapter 2) will be followed to ensure the validity of data in storage, analysis, reporting and archiving. Raw data (including standardized forms and logbooks) are retained permanently in an organized fashion in accordance with EPA records management policies.

Table 5.2-6. Data validation quality control: soils indicator

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

5.3 Hydrology

5.3.1 Introduction

Wetland hydrology is a key driver of wetland ecosystem formation and persistence. Hydrology influences wetland soil condition as well as biotic community composition and structure. In turn, hydrology is controlled by watershed characteristics, and geomorphic conditions found at each site (Tiner 1999, Mitsch and Gosselink 2007). Examples of the types of data to be collected are:

- Degree of saturation
- Degree of inundation
- Types of hydrologic alteration

For more detailed information please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

5.3.2 Sampling Design

The collection of hydrologic data for the NWCA will be entirely in the field - No hydrology samples will be collected for laboratory analysis. Hydrologic data collection is comprised of a number of tasks, including assessment of:

1. hydrologic sources;
2. surface water connectivity to a floodplain;
3. indirect evidence of hydroperiod;
4. hydrologic fluctuations based on evidence of seasonal water levels; and
5. extent of alterations or stressors.

After the Point has been identified, the AA perimeter defined, and the Veg Team has delineated the Vegetation plots (See Section 5.1 above), the AB Team will collect hydrological information from the entire AA. Hydrologic assessment of the Vegetation Plot should be done from the Plot periphery. Groundwater depth information will be collected at the four pits dug to collect soil indicator samples & information. Section 9 of the FOM details how the four pits will be located. Chapter 10 of the FOM details hydrology data collection protocols, as well as the required equipment and standardized data forms.

5.3.3 Sampling and Analytical Methods

All field measurement and observation operations will be conducted by the AB Team.

All observations are recorded on standardized forms which are later entered in to the central NARS surface waters information management system at WED-Corvallis. The form used to collect most Hydrology information is Form H-1. Generally, the AB Team will collect hydrologic information using the following steps:

1. Walk the perimeter of the AA and identify water sources for the AA.
2. Locate the deepest ditch that may provide connectivity between the AA and a floodplain and measure its depth.
3. Search for drift lines and record findings.
4. Use the data form to identify and record any hydrologic alterations found present in the AA including (but not limited to) damming features, ditches and their lengths and depths, and any fresh sediment influx across the wetland.
5. Maximum water depth and the percent of the AA covered by water are recorded on Form WQ-1.
6. At the end of the day just prior to filling in the 4 soil pits, measure the distance from the soil surface down to the surface of the groundwater (recorded on Form S-1).

Annual hydroperiod is covered under other indicator protocols.

Table 5.3-1. Field measurement methods: hydrology metrics.

Variable or Measurement	Units		Summary of Method	
For each pit				
Water Sources			Count of seasonal and perennial sources, including inlets, streams, springs, the ocean, ditches, and pipes	
Hydrologic alterations			Count of damming features (e.g., dikes/berms, roads), length and depth of ditches/drains, evidence of tilling and fresh sediment influx	
Drift lines			Evidence of leaf packs and other plant detritus, anthropogenic trash, and the percent of the AA with standing water.	
Connectivity			Determine the width and depth of the deepest ditch in the AA.	
Water Depth	cm		Determine the maximum depth of surface water and the percent of the AA covered. (Form WQ-1)	
Depth to Groundwater	cm		Recorded on S-1 Form	

5.3.4 Quality Assurance Objectives

As mentioned above in section 2.2.2 (Precision, Bias, and Accuracy), precision of field measurements will not be monitored during the NWCA. Previous hydrology experience or class

work will be valuable for AB team members, but mandatory NWCA training will provide an understanding of basic hydrology. This training will prepare the crew to accurately complete hydrology data collection tasks according to the standardized field protocols.

MQOs are given in Table 5.3-2. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.3-3 represent the maximum allowable criteria for statistical control purposes. Precision is determined from results of the revisits (field measurements) taken on a different day (at least 2-4 weeks apart).

Table 5.3-2. Measurement quality objectives: soil indicator

Variable or Measurement	Precision	Accuracy	Completeness
Field Measurements and Observations	±10%	NA	90%
NA = not applicable in most cases. This would apply if the field auditor did a separate assessment and compared the results to the crews.			

5.3.5 Quality Control Procedures

To avoid impairing data collection for the vegetation indicator, the AB Team members must avoid stepping into the Vegetation Plot modules and potentially trampling vegetation. Assessments of the Vegetation Plot modules should be done from the Plot periphery.

Upon completion of data collection, the Field Crew Leader reviews all forms for completeness and legibility.

In addition, quality assurance audits are conducted, at least once during the field season, for a random subset of field crews to ensure that the protocols are being implemented consistent with training. In addition, ten percent of all sites will receive a repeat visit to determine if differences exist in field data collection on different days.

5.3.6 Data Management, Review, and Validation

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.3-3. The Field Crew Leader is responsible for the validity of all field-generated data (i.e. measurement and observation data) up to the point they are sent to EPA (WED-Corvallis). EPA/ORD QA SOPs (see Chapter 2 for details) will be followed to ensure that data generated and delivered to EPA are valid. Once data have been delivered to EPA, DQ procedures (as detailed in Chapter 2) will be followed to ensure the validity of data in storage, analysis, reporting and archiving. All raw data (including all standardized forms and logbooks) are retained permanently in an organized fashion in accordance with EPA records management policies.

Table 5.3-3: Data quality control: hydrology

Quality Control Activity	Frequency	Acceptance criteria	Corrective Action
Quality Control			
Check completeness of hydrology data	Across AA and Buffer	Values where appropriate	Repeat observations

5.4 Water Chemistry Indicator

5.4.1 Introduction

Along with vegetation and soil, water is one of the key determinants of wetland systems. Some studies show that water chemistry analyses are useful for evaluating wetland ecological integrity and for evaluating stressor-response relationships (Lane and Brown, 2007; Reiss and Brown, 2005). Examples of the types of data to be collected are:

- pH;
- Nutrient Enrichment;
- Dissolved oxygen; and
- Temperature

For more detailed information please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

Water chemistry information will be obtained by collecting samples of surface water for laboratory analysis. At each site, crews fill one 1 L container with surface water. All 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.

5.4.2 Field Collection

While the AA boundary and subdivisions are determined by the Veg Team, the AB Team will determine if surface water meeting the collecting criterion (2x depth of collecting dipper ~ 15 cm) is present within the AA. If there is surface water meeting this criterion, the AB Team will sample the surface water.

5.4.3 Sampling and Analytical Methods

Surface Water Sample and Data Collection:
At the identified sample collection location[6], rinse the collection cup and 1L cubitainer three times, and then collect enough surface water to just fill the 1L cubitainer. Detailed procedures

[6] The preferred sample location will be towards the center of the water body, away from inlets and outlets and deep enough to avoid fouling the water as the dipper is used to collect water

for sample collection and handling are described in the Field Operations Manual, Water Quality Chapter (Sampling Procedure).

Example of long handled dipper in use.
(Photo credit- Maine Dept. of Environmental Protection:
Protocols for Collecting Water Grab Samples in
Rivers, Streams, and Freshwater Wetlands)
http://www.maine.gov/dep/blwq/docmonitoring/biomonitoring/materials/sop_watergrab.pdf

Analysis:
A performance-based methods approach is being utilized for water chemistry 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 for each target analyte, as long as they are able to achieve the performance requirements as listed in Table 5.5-1.

5.4.4 Quality Assurance Objectives

Measurement quality 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.

Table 5.4-1: Performance requirements for water chemistry 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%
pH	pH units	3.7 to 10	NA	NA	5.75 and>8.25	± 0.08 or ± 0.15	± 0.05 or ± 0.10
Ammonia (NH_3)	mg N/L	0 to 17	0.01	0.02[3]	0.10	± 0.01 or ±10%	± 0.01 or ±10%
Nitrate-Nitrite (NO_3-NO_2)	mg N/L	0 to 360 (as nitrite)	0.01	0.02[3]	0.10	± 0.01 or ±10%	± 0.01 or ±10%
Total Nitrogen (TN)	mg N/L	0.1 to 90	0.01	0.02[3]	0.10	± 0.01 or ±10%	± 0.01 or ±10%
Total Phosphorus (TP)	µg P/L	0 to 22,000	2	4[3]	20	± 2 or ±10%	± 2 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 (eq. 1a) as a one-sided 99% confidence interval from repeated measurements of a low-level standard across several calibration curves, and includes medium or mean method blank results, (USGS Open File Report 99-193, EPA 2004). 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 needs 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/ fractional spike recovery, following USGS Open File Report 99-193 and EPA 2004.

4 Value at which performance objectives for precision and bias switch from absolute (≤ transition value) to relative (> 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.

5.4.5 Quality Control Procedures: Field Operations

Control measures to minimize measurement error among crews and sites include the use of standardized field protocols, consistent training of all crews, and availability of experienced technical personnel during the field season to respond to site-specific questions from field crews. Additionally, field crews will apply a consistent labeling convention across all samples (see FOM Water Quality Chapter for details on info to include on labels).

Water chemistry sample duplicates will be collected for performing QA checks. Crews are required to collect a duplicate sample for each 10 surface water samples collected overall. Each crew should collect the QA sample for the first site visited containing sampleable surface water in the AA and then every 10^{th} surface water collection thereafter. This will ensure that a duplicate sample is collected by each crew.

Other controls include audits and revisits. Quality assurance audits are conducted of each Field Crew at least once during the field season, to ensure the protocols followed are consistent with training. Ten percent of all sites will receive repeat sampling visits to be sampled by a Field Crew to determine the extent to which the population estimates might vary if they were sampled at a different time.

Whenever possible, surface water samples should be collected prior to 11:00 a.m. to standardize the collection time frame for the NWCA. This will limit the impact of diurnal changes in the metabolic activity of the organisms in the water. Throughout the water chemistry sample collection process it is important to take precautions to avoid contaminating the sample. Samples can be contaminated quite easily by perspiration from hands, sneezing, smoking, suntan lotion, insect repellent, fumes from gasoline engines or chemicals used during sample collection. Also, the sampler should not enter the water to avoid fouling the water and potentially contaminating or otherwise compromising the quality of the sample. Bottom sediments should not be disturbed as the dipper cup is rinsed three times. The rinse water is poured onto the wetland away from the collection site so that the water does not drain back into the sample area and potentially affect the collected sample. Further, surface water samples should be obtained from areas which are completely free of surface debris.

5.4.6 Quality Control Procedures: Laboratory Operations

5.4.6.1Sample 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 is notified of sample receipt and any associated problems as soon as possible after samples are received. The general schemes for processing wetland water chemistry samples for analysis is presented in Figure 5-4. Several aliquots are prepared from bulk water samples and preserved accordingly. 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 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).

Table 5.4-2. Sample processing quality control activities: water chemistry indicator

Quality Control Activity	Description and Requirements	Corrective Action
Sample Storage	Store samples in darkness at 4°C Monitor temperature daily	Qualify sample as suspect for all analyses
Holding time	Complete processing bulk samples within 48 hours of collection if possible, or ASAP after receipt	Qualify samples
Aliquot Containers and Preparation	HDPE bottles. Rinse bottles and soak for 48 h with ASTM Type II reagent water; test water for conductivity Prepare bottles to receive acid as preservative by filling with a 10% HCl solution and allow to stand overnight. Rinse six times by filling with deionized water. Determine the conductivity of the final rinse of every tenth bottle. Conductivity must be < 2 μS/cm.	Repeat the deionized water rinsing procedure on all bottles cleaned since the last acceptable check. Check conductivity of final rinse on every fifth bottle.
Filtration	0.4 μm polycarbonate filters required for all dissolved analytes. 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.	

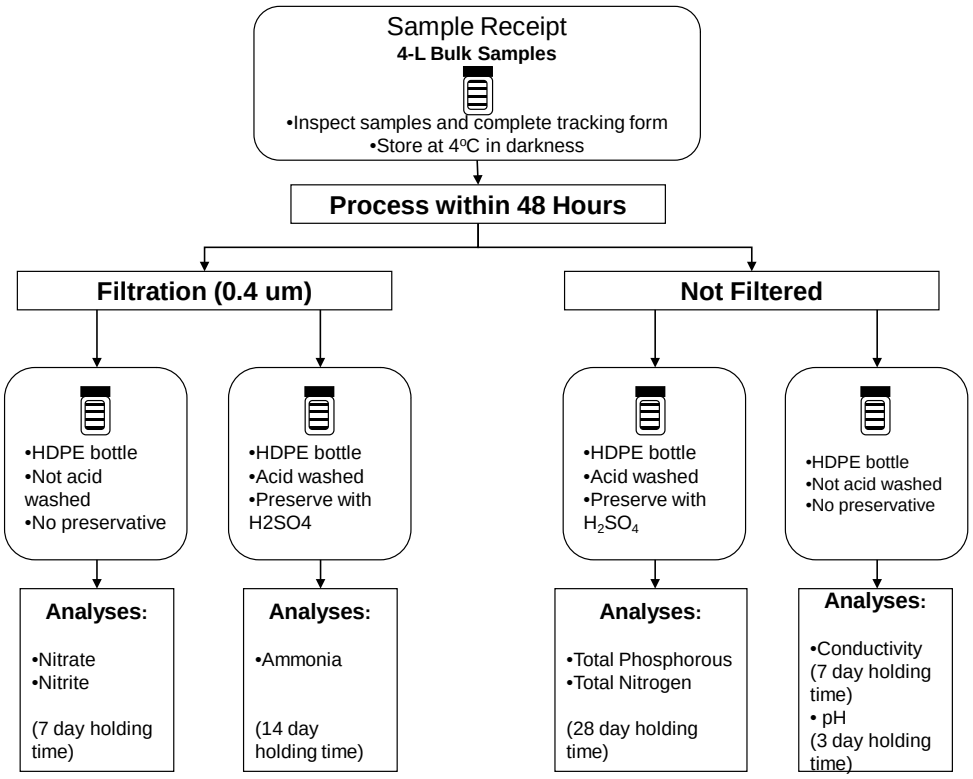


Figure 5-3: General Batch Water Sample Processing Scheme

5.4.6.2Analysis 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. Information regarding QC sample requirements and corrective actions are summarized in Table 5.4-4. Figure 5-5 illustrates the general scheme for analysis of a batch of water chemistry samples, including associated QC samples.

Table 5.4-3. Laboratory Quality Control Samples: Water Chemistry Indicator

QC Sample Type (Analytes), and Description	Frequency	Acceptance Criteria	Corrective Action
Laboratory/ Reagent Blank	Once per day prior to sample analysis	Control limits ≤ LRL	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, ASTM Type II reagent water processed through filtration unit.	Prepare once per week and archive Prepare filter blank for each box of 100 filters, and examine the results before any other filters are used from that box.	Measured concentrations <LDL	Measure archived samples if review of other laboratory blank information suggest source of contamination is sample processing.
LT-MDL Quality Control Check Sample (QCCS) Prepared so concentration is four to six times the LT-MDL objective.	Once per day	Target LT-MDL value	Confirm achieved LRL by repeated analysis of LT-MDL QCCS. Evaluate affected samples for possible re-analysis.
Laboratory Duplicate Sample: (All analyses)	One per batch	Control limits < precision objective	If results are below LRL: 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.

Table 5.4-4. (Continued).

QC Sample Type (Analytes), and Description	Frequency	Acceptance Criteria	Corrective Action
Standard Reference Material: (When available for a particular analyte)	One analysis in a minimum of five separate batches	Manufacturers certified range	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.
Matrix spike samples: (Only prepared when samples with potential for matrix interferences are encountered)	One per batch	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.

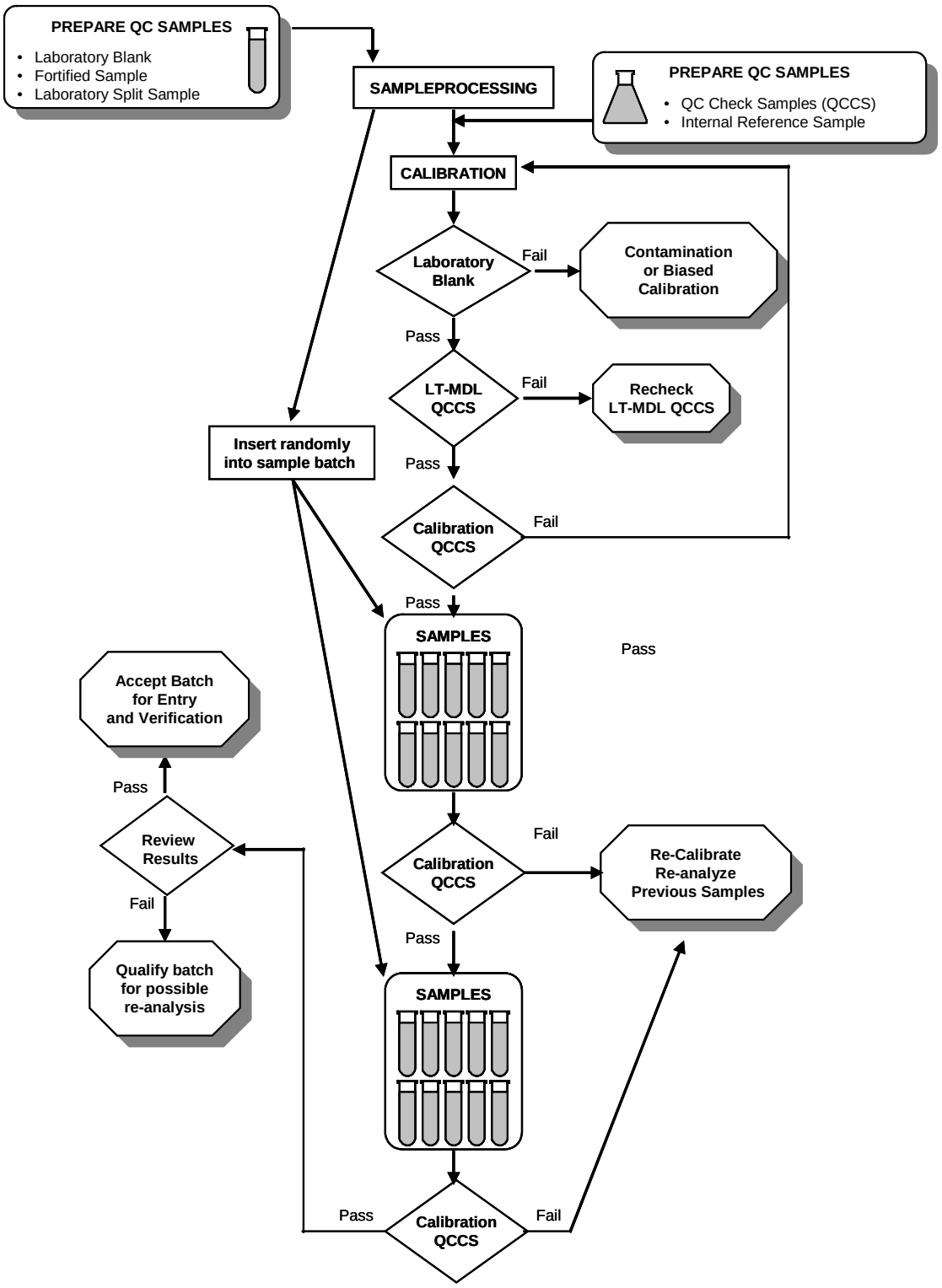


Figure 5-4: Analysis Activities for Water Chemistry Samples

5.4.7 Data Reporting, Review, and Management

Checks made of the data in the process of review and verification are summarized in Table 5.4-5. Data reporting units and significant figures are given in Table 5.4-6. The Project Lead is ultimately responsible for ensuring the validity of the data, although performance of the specific checks may be delegated to other staff members.

Table 5.4-4: Data 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
Review data from QA samples (laboratory PE samples, and interlaboratory comparison samples)	Determine impact and possible limitations on overall usability of data

Table 5.4-5. Data Reporting Criteria: Water Chemistry Indicator

Measurement	Units	No. Significant Figures	Maximum No. Decimal Places
Dissolved Oxygen	mg/L	2	1
Temperature	°C	2	1
pH	pH units	3	2
Conductivity	μS/cm at 25 °C	3	1
Ammonia	mg N/L	3	2
Nitrate-Nitrite	mg N/L	3	2
Total nitrogen	mg N/L	3	2
Total phosphorus	μg P/L	3	0

5.5 Algae Indicator

5.5.1 Introduction

Algae, which include planktonic (open water), benthic (periphyton), and metaphyton forms, are an extremely important ecosystem component, providing essential primary productivity as well as being a food resource for higher level organisms including macroinvertebrates and fish (Mitsch and Gosselink 2007). Like other biotic taxa, many indicator attributes or metrics which describe ecological condition can be derived from data describing taxonomic composition and abundance of algae. Due to high dispersal and growth rates, algae respond quickly to environmental disturbances, both natural and anthropomorphic, and are one of the first

indicators of ecological change in the wetland (McCormick and Cairns 1994). Examples of the types of data to be collected are:

- Species composition and abundance, including guilds
- Productivity
- Toxicity

For more detailed information please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

5.5.2 Sampling Design

Algae collection procedures for the NWCA have generally been based upon the multi-habitat procedures of the National Water-Quality Assessment Program (NAWQA) (Moulton et al 2002). The design is based on a representative multi-habitat or composite sampling method, thus samples are collected from multiple habitats rather than once at the POINT. This method provides a qualitative sample, though a fixed area of sampling effort is used. Multi-habitat samples will be collected from surface sediments (benthic sample), and from the surface of vegetation stems or leaves if vegetation is present. Multi-habitat samples collected at the site are composited in a bottle and homogenized to characterize taxonomic composition and relative abundance of the algal assemblage in the AA. If the wetland AA has standing water present, a phytoplankton sample will also be collected for biomass estimates.

A 250 ml sub-sample of the composite sample is collected and shipped to the designated lab for identification. Surface water and epiphytic algae composite samples are sub-sampled to test for microcystin toxicity. The phytoplankton sample for biomass (chlorophyll a) analysis is collected on a glass fiber filter and shipped to the designated lab.

It is anticipated that the AB Team will collect and field process all algae samples. At the end of the day's sampling activities, at least one type of laboratory sample (i.e. to evaluate composite taxonomic composition) will have been taken at all sites. In addition, a biomass sample will be collected at all sites with sampleable surface water.

5.5.3 Sampling and Analytical Methods

Three distinct types of samples will be collected: composite taxonomic samples, and, if the AA includes standing water, a toxin sample and a biomass (chlorophyll a) sample. The toxin sample will consist of five epiphytic algae samples and surface water and the composite taxonomic sample will include the five epiphytic algae samples and surface water, as well as five substrate samples. All the algae sampling will be conducted by the AB Team. While the AA is determined by the Veg Team, the AB Team will determine algal sample collection locations in the AA for taxonomic assemblage, as well as determine if surface water meeting the collecting criterion (2x depth of collecting dipper ~ 6 inches) is present within the AA.

If standing water is located in the vegetation plot only, the AB Team will work with the Veg Team to minimize vegetation impacts while collecting the algal sample.

5.5.3.1Microcystin Toxin Sample

Sample Collection:
If aquatic or emergent plants are present, epiphytic algae samples are collected from one inch square surface area plant scrapings for a total of 5 samples per AA. These are rinsed into a 125 mL bottle and surface water is used to fill it to the shoulder. Fifty mL of this sample are measured out and used for the taxonomic ID sample, as described below, and surface water, is again added to the shoulder of toxin sample. If no epiphytes are present, the bottle is simply filled to the shoulder with surface water. The sample is then properly labeled and stored and shipped on ice.

Lab Analysis:
Toxin samples will be processed by performing Microtiter Plate ELISA of Microcystins using the Abraxis Polyclonal ADDA kit at the USGS Organic Geochemistry Research Laboratory (OGRL) in Lawrence, KS. Results for water samples and concentrations are reported between 0.10 µg/L and 5.0 µg/L without dilution.

5.5.3.2Composite Taxonmic Sample

Sample Collection if no surface water:
If there is no evidence of previous inundation or desiccated algae, a sample is not collected. If suitable substrate is located with probable algal growth, ten substrate samples are collected using a sampling device described by Moulton et al 2002. Sediment core samples are collected using a 1-inch square modified syringe. The target length for a core sample is the top few millimeters (1/2-inch). If the target length cannot be obtained after two consecutive attempts, the maximum obtainable core should be used. All samples will be deposited into a 250 mL bottle which will be filled to the shoulder with deionized water, then homogenized. Lugols is added to preserve the sample.

Sample Collection if AA has surface water:
Two types of surfaces are selected within the AA that are suitable for sampling including depositional habitats (soft bottom, stones, sticks/wood) and vegetation for epiphytic habitat. Fifty mLs of the algal toxin sample is poured into the 250 mL taxonomic ID bottle. This provides the the portion needed of epiphytic algae. Five more samples are collected and added to the bottle. These samples are representative of the predominant surfaces in the water being sampled. One inch square surface area scrapings or ½-in core samples are collected from each microhabitat, then the bottle is filled to the shoulder with surface water. This will make a total of 10 samples per AA, which are homogenized and preserved with Lugols. Detailed procedures for sample collection and handling are described in chapter 9 of the Field Operations Manual.

Lab Analysis:
Sediment samples are cleaned of organic matter with strong oxidizing agents and slides are made. The analysis is made by identifying and counting 600 individual cells. Detailed procedures for sample processing and enumeration are described in the laboratory methods manual. Table 5.5-1 summarizes field and analytical methods for the diatoms and Table 5.5-2 summarizes the field an analytical methods for soft algae.

Table 5.5-1. Field and laboratory methods: Diatoms

Variable or Measurement	QA Class	Expected Range and/or Units	Summary of Method	References
Sample Collection	C	NA	Core sampler used to collect a 1 cm core of sediments, epiphytic algae or algae from other substrates. Surface water is also added	Glew et al. 2001; Wetlands Survey Field Operations Manual 2009
Sample Digestion and Concentration	N	NA	Add acid and heat at 200°C for 2 hrs. Allow to settle, siphon off supernatant, repeat until final volume is between 25-50 mL	Charles et al. 2003; Wetlands Survey Laboratory Methods Manual 2010
Slide preparation	N	NA	Prepare coverslips and mount on slide using Naphrax	Charles et al. 2003; Wetlands Survey Laboratory Methods Manual 2010
Enumeration	C	0 to 300 organisms	Random systematic selection of rows and fields with target of 600 organisms from sample	Charles et al. 2003; Wetlands Survey Laboratory Methods Manual 2010
Identification	C	genus	Specified keys and references	

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

Table 5.5-2. Field and laboratory methods: Soft Algae

Variable or Measurement	QA Class	Expected Range and/or Units	Summary of Method	References
Sample Collection	C	NA	Core sampler used to collect a 1 cm core of sediments, epiphytic algae or algae from other substrates. Surface water is also added	Glew et al. 2001; Wetlands Survey Field Operations Manual 2009
Plamer-Maloney Cell Preparation	N	NA	0.05mL of soft algae subsample viewed in two, half Plamber-Maloney cells	USGS 1997; NAWQA Laboratory Methods Manual
Sedgewick-Rafter Cell Preparation	N	NA	Large soft algae viewed in Sedgewick-Rafter cell	USGS 1997; NAWQA Laboratory Methods Manual
Enumeration	C	0 to 300 organisms	Random systematic selection of rows and fields with target of 600 organisms from sample	USGS 1997; NAWQA Laboratory Methods Manual
Identification	C	genus	Specified keys and references	

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

5.5.3.3Biomass (chlorophyll a) Sample

Sample Collection:
Water samples are collected using the long-handled dipper from the water chemistry protocol. Take care to minimize disturbance of submerged, floating or emergent vegetation and associated epiphytes so that none of this material is collected. Also, do not sample duckweed, *Wolfia*, etc. 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 FOM Algae Chapter.

Lab 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 5.5-3.

Table 5.5-3. Performance Requirements for *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]
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 (eq. 1a) as a one-sided 99% confidence interval from repeated measurements of a low-level standard across several calibration curves, and includes medium or mean method blank results, (USGS Open File Report 99-193, EPA 2004). 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 needs 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/ fractional spike recovery, following USGS Open File Report 99-193 and EPA 2004.

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.

5.5.4 Quality Assurance Objectives

5.5.4.1Composite Taxonomic Sample

A taxonomic harmonization table for diatoms will be developed through co-operation of the different taxonomic laboratories to ensure consistent identification among laboratories. The harmonization table will begin with the National Water-Quality Assessment (NAWQA) program diatom list, and taxonomic experts from each laboratory will work together to clean up the data set to ensure that there will be no ambiguous or synonymous taxa in the final data set.

5.5.4.2 Microcystin Toxin Sample and Biomass (chlorophyll a) Sample:

MQOs are given in Table 5.5-3. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.5-3 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 5.5-3 represent the 99 percent 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 percent 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, including those in Table 5.5-4 (field) and Table 5.5-5 (lab).

5.5.5 Quality Control Procedures: Field Operations

5.5.5.1 Composite Taxonomic Sample and Mycrocystin Toxin Sample:

Any contamination of the samples can produce significant errors in the resulting interpretation. Great care must be taken by the samplers not to contaminate the bottom sample with higher levels of the core or with surface water or with the tools used to collect the sample (i.e., the corer, core tube, and spatulas). Prior to sampling, the corer device and collection tools should be examined to ensure that they are clean and free of contaminants from previous sampling activities. After the core is sectioned off, the sectioning apparatus should be removed and rinsed in DI water.

After each sample is placed in the container, the label 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 sample is recorded correctly on the Sample Collection Form (Figure 4-4). 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.

5.5.5.2Biomass (chlorophyll a) Sample:

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 incremental smaller volumes.

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 (Figure 5-6). 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.

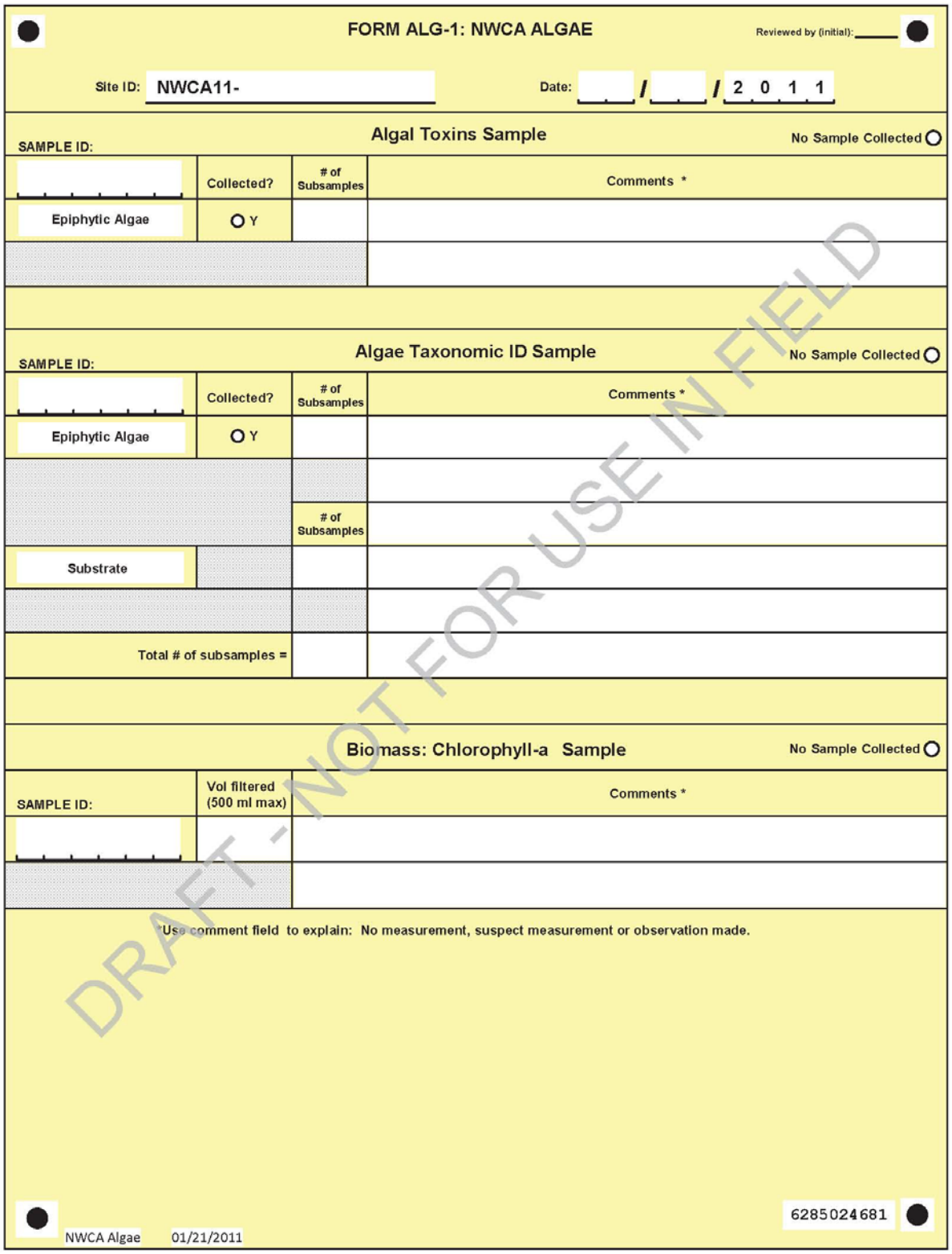
Table 5.5-4. Field Sample Processing Quality Control: *chlorophyll a* Samples

Quality Control Activity	Description and Requirements	Corrective Action
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

Wrap the vial in foil to keep sunlight from degrading the sample and place the vial in a small Whirl-Pak bag also labeled with Site_ID, Date, and chlorophyll a. Immediately place the sample in an ice chest filled with ice or dry ice.

Record the *chlorophyll a* sample data on the data log sheet along with other perishable sample's data going into the ice chest.

Send the sample to the contracted lab overnight via FedEx. If the *chlorophyll a* samples are held for a period prior to shipping, place the samples in a freezer until they can be shipped.



FORM ALG-1: NWCA ALGAE

Reviewed by (initial): ____

Site ID: NWCA11- Date: ___ / ___ / 2011

Algal Toxins Sample — No Sample Collected ○

SAMPLE ID:	Collected?	# of Subsamples	Comments *
Epiphytic Algae	○ Y		

Algae Taxonomic ID Sample — No Sample Collected ○

SAMPLE ID:	Collected?	# of Subsamples	Comments *
Epiphytic Algae	○ Y		
		# of Subsamples	
Substrate			
Total # of subsamples =			

Biomass: Chlorophyll-a Sample — No Sample Collected ○

SAMPLE ID:	Vol filtered (500 ml max)	Comments *

*Use comment field to explain: No measurement, suspect measurement or observation made.

DRAFT - NOT FOR USE IN FIELD

NWCA Algae 01/21/2011 6285024681

Figure 5-5: Sample Collection Form

5.5.6 Quality Control Procedures: Laboratory Operations

5.5.6.1Composite Taxonomic Sample

A total of 10% of the samples collected will be analyzed for quality control. Analysts will swap vials of material and recount only the dominant taxa (10-15% or more of the units counted). Re-counts will be performed by another experienced taxonomist at an independent laboratory who did not participate in the original identifications. EPA will inform the laboratories which random samples will be re-counted. The samples must then be sent from the original laboratory to the independent laboratory. The QC taxonomist should complete another copy of the Taxonomic Bench Sheet for each sample. Each bench sheet should be labeled with the term "QC Re-ID." As each bench sheet is completed, it should be faxed to the Information Management Coordinator.

EPA will compare the taxonomic results generated by the primary and QC taxonomists for each sample based on both the raw data and the appropriate metrics (i.e., taxa identified with similar autecologies). EPA will then calculate percent similarity. It is expected that the soft algae counts should have a similarity of ≥50% and the diatom counts should have a similarity of ≥70%. If not, the reasons for the discrepancies between taxonomists should be discussed. Results less than these values will be investigated and logged for indication of error patterns or trends.

A report or technical memorandum will be prepared by the QC taxonomists. This document will quantify both aspects of taxonomic precision, assess data acceptability, highlight taxonomic problem areas, and provide recommendations for improving precision. This report will be submitted to the Information Management Coordinator, with copies sent to the primary and QC taxonomists and another copy maintained in the project file. Significant differences may result in the re-identification of samples by the primary taxonomist and a second QC check by the secondary taxonomist. All samples must be stored at the laboratory until the project officer notifies the lab.

5.5.6.2Microcystin Toxin Sample

The Quality Assurance Officer or designee will evaluate overall data quality and QC compliance. In the event data is not in compliance, the problem(s) will be identified and samples will be reanalyzed, as appropriate, after corrective action is taken.

The standard curve should have a correlation coefficient of 0.99 (as suggested by ELISA kit manufacturer).

The absorbency of the blank must be >1.400 (as suggested by ELISA kit manufacturer).
The Check Standard supplied with the ELISA kit should be analyzed a minimum of two times in each run. Once at the beginning and once at the end. This helps ensure the plate was prepared in the proper time frame. Values should be +/- 20 % (28.3% relative standard deviation (RSD)) of expected value.

Laboratory duplicates should have a percent Relative Standard Deviation (% RSD) of 28.3 percent or less when compared to each other (as suggested by ELISA kit manufacturer).
If laboratory duplicates are outside of this range, then they should be reanalyzed in the next run.

Quality control samples are available for each project. Criteria for acceptance of measured values are +/- 20% of expected concentration. These samples are analyzed every time samples from the same project code are analyzed.

A designated archived project sample is reanalyzed with every run that is analyzed. Control charts are maintained for these samples. A running historical average is maintained of the concentration from each run. The concentration of the QC sample for each successive run has to be ± 20 percent of that average to be acceptable.

5.5.6.3Biomass (chlorophyll a) Sample

QC activities associated with sample receipt and processing are presented in Table 5.5-5. The communications center and information management staffs are notified of sample receipt and any associated problems as soon as possible after samples are received.

Table 5.5-5. Sample Processing Quality Control: Composite and *chlorophyll a* Samples

Quality Control Activity	Description and Requirements	Corrective Action
Sample Storage	Store samples in darkness and frozen (-20 °C) Monitor temperature daily	Qualify sample as suspect for all analyses

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, where applicable, and corrective actions are summarized in Table 5.5-6.

Table 5.5-6: Lab sample processing quality controls: *chlorophyll a*.

QC Sample Type (Analytes), and Description	Frequency	Acceptance Criteria	Corrective Action
Laboratory Duplicate Sample: (All analyses)	One per batch	Control limits < precision objective	If results are below LRL: 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.

Standard Reference Material: (When available for a particular analyte)	One analysis in a minimum of five separate batches	Manufacturers certified range	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.
Matrix spike samples: (Only prepared when samples with potential for matrix interferences are encountered)	One per batch	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.

5.5.7 Data Management, Review, and Validation

5.5.7.1Composite Taxonomic Sample

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.5-7. The Project 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 NWCA project. The electronic data files are transferred to NWCA 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 NWCA 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 in an organized fashion by the IM Staff permanently or until written authorization for disposition has been received from the EPA Project Leader.

Table 5.5-7. Laboratory Quality Control: Composite Sample (Diatoms and Soft Algae)

Check or Sample Description	Frequency	Acceptance Criteria	Corrective Action
IDENTIFICATION			
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 standard 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 Leader
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
DATA VALIDATION			
Taxonomic "reasonable-ness" checks	All data sheets	Genera known to occur in given site or geographic area	Second or third identification by expert in that taxon

5.5.7.2 Microcystin Toxin Sample and Biomass (chlorophyll a) Sample:

Checks made of the data in the process of review, verification, and validation are summarized in Table 5.5-8. Data reporting units and significant figures are given in Table 5.5-9. The Project 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 NWCA project. The electronic data files are transferred to the NWCA 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 NWCA IM Coordinator.

Table 5.5-8. Data 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 5.5-9. Data reporting criteria: *chlorophyll a* indicator

Measurement	Units	No. Significant Figures	Maximum No. Decimal Places
chlorophyll a	µg/L	2	1

5.6 Stressors Indicator

Stressors are an important component of an assessment of wetlands because they degrade ecological condition and can be used to determine management responses designed to improve condition (Adamus and Brandt 1990). As the number of stressors accumulates, it is assumed overall wetland condition declines. We also assume this relationship holds true regardless of wetland class. All or most of the data collection of stressors will be done as part of the protocols of the other indicators. For example, hydrology-related stressors will be addressed under the hydrology indicator in the "NWCA Field Operations Manual" (EPA- 843-R-10-001) For more detailed information on stressors and their application to wetland assessment please see "Ecological Indicators for the 2011 National Wetland Condition Assessment" (in preparation).

5.7 Rapid Assessment Method

5.7.1 Introduction

The primary purpose of the USA-RAM is to assess overall condition and stress for the nation's wetlands as part of the NWCA. The secondary purposes is to provide a rapid assessment method to U.S. states and Tribes that they can further develop for their own purposes.

USA-RAM focuses on the form and structure of wetlands. For any wetland class, we assume that wetland with more complex form and structure, and less stress, tends to support higher levels of ecological integrity.[7] Individual metrics within the condition index are selected and organized to reflect a set of four core wetland attributes describing ecosystem structure and form. One attribute reflects wetland hydrology as represented by water level fluctuation and connectivity to the other aquatic resources. Another attribute reflects physical structure as represented by topographic complexity and patch mosaic complexity in a wetland assessment area. The third attribute is biological structure of the wetland is expressed in terms of the vertical complexity of the vegetation community and overall plant community complexity. A fourth attribute termed buffer is also part of the condition index.

The presentation of stressor metrics within USA-RAM is based on an assessment framework that assumes wetland exposure to anthropogenic disturbance will affect ecosystem condition[2]. The magnitude of those effects is related to the proximity, intensity and duration of stressors acting on the wetland in a cumulative way. These influences and their interactions cannot be assessed with a known level of certainty using USA-RAM. Instead, USA-RAM relies on an approach that classifies the number of human caused stressors that cause wetland degradation. The overall stress on a wetland is assessed as the number of evident stressors and their

[7] Ecological Integrity: The condition of an unimpaired ecosystem as measured by combined chemical, physical (including physical habitat) and biological attributes.
[2] Ecological Resilience: The capacity of an ecosystem to withstand disturbance and human-induced stress.

intensity. As the number of stressors accumulates, wetland overall condition declines. We assume that this relationship holds true regardless of wetland class.

5.7.2 Sampling Design

USA-RAM is designed to assess overall condition and stress for a 0.5-ha circular Assessment Area (AA). Condition and stress are assessed separately for each of four attributes (Buffer, Hydrology, Physical Structure, and Biological Structure), based on unique metrics and their field indicators. The same attributes, metrics, and indicators are applied to every AA. Details on the field protocol can be found in USA-RAM Manual (Collins and Fennessy 2010).

Attributes	Condition Metrics	Stress Metrics
Buffer	Percent of AA Having Buffer	Stress to the Buffer Zone
	Buffer Width	
Hydrology	Water Level Fluctuation	Stressors to Water Quality
	Hydrological Connectivity	Alterations to hydroperiod
Physical Structure	Topographic Complexity	Habitat/Substrate Alterations
	Patch Mosaic Complexity	
Biological Structure	Vertical Complexity	Percent Cover of Invasive Plants
	Plant Community Complexity	Vegetation Disturbance

This rapid assessment method uses presence/absence checklist and other semi-quantitative and narrative metrics that rely on best professional judgment and onsite evidence to measure aspects of landscape, hydrology, physical structure, and biological structure to generate individual attribute and aggregate scores to reflect condition on the site.

No USA-RAM data will be sent to a laboratory for further analysis; all metrics are based on field observations.

5.7.3 Quality Assurance Objectives

MQOs are given in Table 5.7-1. General requirements for comparability and representativeness are addressed in Section 2. The MQOs given in Table 5.1-4 represent the maximum allowable criteria for statistical control purposes. Precision is determined from results of revisits (field measurements) taken on a different day (at least 2-4 weeks apart).

Table 5.7-1. Measurement data quality objectives: vegetation indicator

Variable or Measurement	Precision	Accuracy	Completeness
Field Measurements and Observations	±10%	NA	90%
NA = not applicable in most cases. This would apply if the field auditor did a separate assessment and compared the results to the crews.			

5.7.4 Quality Control Procedures: Field Operations

Precision, bias and accuracy of field measurements will not be monitored during the NWCA[8]. Control measures to minimize measurement error among crews and sites include the use of standardized field protocols, consistent training of all crews, field assistance visits to all crews, and availability of experienced technical personnel during the field season to respond to site-specific questions from field crews as they arise.

USA-RAM data is collected independently from other NWCA field data to allow for un-biased calibration of the USA-RAM based on the more intensive NWCA data. The Field Crew Leader directs the AB Team to complete all USA-RAM sampling activities upon arriving at the site.

Upon completion of sampling, the Field Crew Leader reviews all USA-RAM forms for completeness, legibility, and errors.

5.7.5 Data Management, Review, and Validation

The Field Crew Leader is responsible for the validity of all field-generated data (i.e. measurement and observation data) up to the point it is sent to EPA (ORD/Corvallis. Once data have been delivered to EPA, DQ procedures (as detailed in Chapter 2) will be followed to ensure the validity of data in storage, analysis, reporting and archiving. All raw data (including all standardized forms and logbooks) are retained permanently in an organized fashion in accordance with EPA records management policies. No USA-RAM data will be sent to a laboratory for further analysis; all metrics are based on field observations.

Tables for scoring each Metric are provided in this version of USA-RAM. The same tables are included in a separate set of data sheets designed for use in the field. These scoring tables are preliminary. After the method is fully tested, the scoring tables will be removed from the manual and the field data sheets. The final data sheets will only include the input data used to calculate the Metric scores. Results from surveys of the regional networks of reference sites will be used to develop Metric scoring tables for each region of the U.S. It is anticipated that the score for each Attribute will be the sum of the scores for its respective Metrics, and that each AA score will be the sum of its Attribute scores. Every AA will have one AA score, a set of Attribute scores, and a set of Metric scores.

[8] Bias, for example, cannot be determined directly, since the “true” values at any particular site are not known.

6 FIELD AND LABORATORY QUALITY EVALUATION AND ASSISTANCE VISITS

No national program of accreditation for vegetation and sample 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 Wetland Condition Assessment.

Procedural review and assistance personnel are trained to the specific implementation and data collection methods detailed in the NWCA: Field Operations Manual (USEPA, 2011[b]). Plans and checklists for field evaluation and assistance visits 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 actions 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. Uniform training of field crews and review cycles conducted early in the data collection process will significantly reduce sampling variability associated with specific implementation or interpretation of the protocols. The field visits 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 and assistance visit 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 as follows:

6.1 Field Quality Evaluation and Assistance Visit Plan for the National Wetland Condition Assessment (NWCA)

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

To Evaluate: Regional Monitoring Coordinator-appointed Field Crews during sampling operations on site.

Purpose: To identify and correct deficiencies during field sampling operations.

1. Training staff will review the Field Evaluation and Assistance Visit Plan and Check List with each Evaluator during field operations training sessions.

2. The Contractor QA Officer or authorized designee will send a copy of the final Plan and 4-part carbonless copy versions of the final Check List pages, envelopes to return the Check Lists, a clipboard, pens, and NWCA QAPP and Field Operations Manual to each participating Evaluator.

3. Each Evaluator is responsible for providing their own field gear sufficient to accompany the Field Crews (e.g., protective clothing, sunscreen, insect repellent, hat, 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 Contractor QA Officer and respective Field Crew Leader. Evaluators should be prepared to spend additional time in the field if needed (see below).

4. TBD Contractor and the Regional Coordinators will arrange the schedule of visitation with each Field Crew, and notify the Evaluators concerning site locations, where and when to meet the Crew, and how to get there. Ideally, each Field Crew 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 and evaluate TBD Contactor Field Crews. Any EPA or Contractor Evaluator may visit State/Tribal Field Crews.

5. An NWCA Field Crew consists of four persons where, at a minimum, the Field Crew Leader is fully trained.

6. If membership of a Field Crew changes, and at least two of the members have not been evaluated previously, the Field Crew must be evaluated again during sampling operations as soon as possible to ensure that all members of the Field Crew understand and can perform the procedures.

7. The Evaluator will view the performance of a Crew 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 Crew to the next site to complete the evaluation of the first activities on the list.

    b. If the Crew 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 Crew Leader before leaving the site (if practicable), noting positive practices and problems (i.e., weaknesses [might affect data quality]; deficiencies [would adversely affect data quality]). The Evaluator will

ensure that the Crew 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. They will review this list with the field sampling crew at the site.

e. If the Evaluator's findings indicate that the Field Crew is not performing the procedures correctly, safely, or thoroughly, the Evaluator must continue working with this Field Crew until certain of the Crew'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 Crew operations (e.g., less than three members, equipment or performance problems) the Evaluator must contact one of the following QA officials:

   i. Regina Poeske, EPA NWCA QA Assistance Visit Coordinator (215) 814-2725.
   ii. Sarah Lehmann, EPA NWCA Project QA Officer (202-566-1183)

   The QA official will contact the EPA Project Leader (Michael Scozzafava – 202-566- 1376) or Alternate EPA Project Leader (Chris Faulkner – 202-566-1185 or Gregg Serenbetz 202-566-1253) to determine the appropriate course of action.

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

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

10. Fasten the pages of the check list for each Field Crew together with a paper clip.

11. Mail the remaining pages of each completed Field Evaluation and Assistance Check List to:

    Marlys Cappaert
    SRA/Raytheon
    USEPA-WED
    200 West 25th St.
    Corvallis, OR 97333

    Each set of Assistance Visit forms will be scanned and the data will be entered into the NWCA Information Management System. The EPA NWCA QA Assistance Visit Coordinator will review the Field Evaluation and Assistance Check Lists, note any issues, check off the completion of the evaluation for each Field Team

6.2 Laboratory Quality Evaluation and Assistance Visit Plan for the National Wetland Condition Assessment (NWCA)

Evaluators: One or more designated NWCA QA Assistance Visit staff members who are qualified (i.e., have completed training) in the procedures of the NWCA laboratory operations.

To Evaluate: Laboratories performing chemical, vegetation, diatom or algal analysis or subsampling, sorting, and taxonomic procedures to analyze wetland samples.

Purpose: To identify and correct deficiencies during laboratory operations and procedures.

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

2. The Contractor QA Officer or authorized designee will send a copy of the final Plan and 4-part carbonless copy versions of the final Check List pages, envelopes to return the Check Lists, a clipboard, pens, and NWCA QAPP and Laboratory Methods manual to each participating Evaluator.

3. Each laboratory analyzing samples will receive an Assistance Visit or equivalent evaluation from an Evaluator. Those laboratories receiving assistance visits include the Natural Resource Conservation Service Laboratory (soil chemistry and bulk density) in Lincoln, Nebraska and EcoAnalyst Laboratory (algae and vegetation taxonomy) in Moscow, Idaho. A remote evaluation procedure will be used for all water chemistry laboratories including the U.S. Geological Survey Laboratory in Denver, CO, the Wisconsin State Laboratory in Madison, WI, and the ORD Western Ecology Division Laboratory in Corvallis, OR. The Evaluator will also use a remote evaluation procedure for all state vegetation laboratories. An interlaboratory comparison investigation will be used for the algae toxins laboratories including the Wisconsin State Laboratory in Madison, WI and the U.S. Geological Survey Kansas Water Science Center in Lawrence, KS.

Lab	Analysis	Type of Evaluation
Natural Resource Conservation Service (NRCS) Lab - Lincoln, NE	Soil Chemistry and Bulk Density	Assistance Visit
EcoAnalyst Lab - Moscow, ID	Algae and Vegetation Taxonomy	Assistance Visit
U.S. Environmental Protection Agency (EPA) ORD Western Ecology Division Lab - Corvallis, OR - Dynamac	Water Chemistry	Remote Evaluation
U.S. Geological Survey - Denver, CO	Water Chemistry	Remote Evaluation
Wisconsin State Labs - Water Chemistry	Water Chemistry	Remote Evaluation
State Vegetation Labs	Vegetation Taxonomy	Remote Evaluation
U.S. Geological Survey - Kansas Water Science Center, Lawrence, KS	Algal Toxin	Interlaboratory Comparison

Wisconsin State Lab - Algal Toxins	Algal Toxins	Interlaboratory Comparison

4. The Contractor will make sure that all lab audits are performed before the first ten percent of samples are analyzed, 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 schedule lab visits, schedule teleconference calls, and obtain documentation in consultation with the Contractor QA Officer and the respective Laboratory Supervisor Staff. Evaluators should be prepared to spend additional time in the laboratory if needed (see below).

    a. For those laboratories (Natural Resource Conservation Service Laboratory and EcoAnalyst Laboratory) receiving an Assistance Visit, the Evaluator will observe the performance of the laboratory procedures and QC Officer through one complete set of sample processing activities as detailed on the Laboratory Evaluation and Assistance Check List.

        i. 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 laboratory personnel when a new sample is started to complete the evaluation of the first activities on the list.

        ii. If laboratory personnel miss or incorrectly perform 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 Laboratory Operations Manual, all data are recorded correctly, and paperwork is properly completed at the site.

        iii. When the sample has been completely processed or analyzed, the Evaluator will review the results of the evaluation with laboratory personnel and QC Officer, noting positive practices and problems (i.e., weaknesses [might affect data quality]; deficiencies [would adversely affect data quality]). The Evaluator will ensure that the laboratory personnel and QC Officer understand the findings and will be able to perform the procedures properly in the future.

        iv. The Evaluator will record responses or concerns, if any, on the Laboratory Evaluation and Assistance Check List. All Laboratory Evaluations and completed checklists are sent to the NWCA Project Manager. The NWCA Project Manager will retain the records permanently in an organized fashion in accordance with EPA records management policies.

        v. 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.

vi. If the Evaluator finds major deficiencies in the Laboratory operations, the Evaluator must contact one of the following QA officials:

1. Regina Poeske, EPA NWCA QA Assistance Visit Coordinator (215) 814-2725.

2. Sarah Lehmann, EPA NWCA Project QA Officer (202-566-1183)

The QA official will contact the EPA Project Leader (Michael Scozzafava – 202-566-1376) or Alternate EPA Project Leader (Chris Faulkner – 202-566-1185 or Gregg Serenbetz 202-566-1253) to determine the proper course of action. Data records from samples previously processed by this Laboratory will be checked to determine if samples must be redone. In cases where irresolvable deficiencies are noted, the EPA Project Leader will direct the laboratory to stop processing samples and send them to another laboratory.

b. For those water chemistry laboratories receiving a remote evaluation (U.S. Geological Survey Laboratory in Denver, CO, the Wisconsin State Laboratory, and the ORD Laboratory), the Evaluator will request the laboratory to provide documentation of its policies and procedures (see Section 1.3.2 Overview of Laboratory Operations and Laboratory SOPs in Chapter 2 for details), including:

i. The laboratory's Quality Manual, Quality Management Plan or similar document

ii. Standard Operating Procedures (SOPs) for each analysis to be performed

iii. Method Detection Limits (MDLs) for each instrument used and Demonstration of Capability (DOC) for each analysis to be performed

iv. A list of the laboratory's accreditations and certifications, if any

v. Results from Proficiency Tests for each analyte to be analyzed under the NWCA project

If a laboratory has clearly documented procedures for sample receiving, storage, preservation, preparation, analysis, and data reporting; has successfully analyzed Proficiency Test samples; has a Quality Manual that thoroughly addresses laboratory quality including standard and sample preparation, record keeping and QA non-conformance; participates in a nationally recognized or state certification program; and has demonstrated ability to perform the testing for which program/project the audit is intended, then the need for an on-site visit will be waived. The EPA Project Leader will make a final decision on the need for an actual on-site visit after the review and evaluation of the documentation requested.

c. For participating state vegetation laboratories that are receiving a remote evaluation, the Evaluator will disseminate a checklist to each participating laboratory and schedule a teleconference to discuss the checklist. This teleconference will also be used as an opportunity for the laboratories to ask questions about the analytical procedures, tracking, and reporting requirements.

i. The role of the Evaluator is to provide additional training and guidance so that the procedures are performed consistent with the Laboratory Operations Manual, all data are recorded correctly, and paperwork is properly completed at the site. For vegetation laboratories, the checklist will focus on how carefully the specimens were pressed, preserved and stored prior to their identification by the expert botanists. During the teleconference, the Evaluator will note any incorrectly performed procedures on the checklist and immediately point them out so the mistake can be corrected.

ii. When the teleconference call is complete, the Evaluator will review the results of the evaluation with laboratory personnel and QC Officer, noting positive practices and problems (i.e., weaknesses [might affect data quality]; deficiencies [would adversely affect data quality]). The Evaluator will ensure that the laboratory personnel and QC Officer understand the findings and will be able to perform the procedures properly in the future. The Evaluator will send an email summary of the evaluation findings to each laboratory. All Laboratory Evaluations and completed checklists are sent to the NWCA Project Manager. The NWCA Project Manager will retain the records permanently in an organized fashion in accordance with EPA records management policies.

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

iv. 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.

v. After all identifications have been completed by each state vegetation laboratory, each state will calculate their Percent Taxonomic Difference. If this value is not less than or equal to fifteen percent, the Evaluator will follow up with the laboratory to make sure they are aware of this deficiency. The EPA NWCA QA Assistance Visit Coordinator will arrange a conference call between the participating laboratory botanists to try to resolve the conflicting identifications.

vi. If the Evaluator finds major deficiencies in the Laboratory operations, the Evaluator must contact one of the following QA officials:

1. Regina Poeske, EPA NWCA QA Assistance Visit Coordinator (215) 814-2725.

2. Sarah Lehmann, EPA NWCA Project QA Officer (202-566-1183)

The QA official will contact the EPA Project Leader (Michael Scozzafava – 202-566-1376) or Alternate EPA Project Leader (Chris Faulkner – 202-566-1185 or Gregg Serenbetz 202-566-1253) 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 reidentified. In cases

where irresolvable deficiencies are noted, the EPA Project Leader will direct the laboratory to stop processing samples and send them to another laboratory.

d. For those laboratories (Wisconsin State Laboratory and the U.S. Geological Survey Kansas Water Science Center) undergoing an interlaboratory investigation, the Evaluator will coordinate a blind interlaboratory comparison for microcystin measurements. This comparison will include an analysis of spiked and unspiked samples by the two participating laboratories. The study design for this interlaboratory comparison is found in Appendix C. No site visit is envisioned for these labs unless the data submitted and reviewed by EPA does not meet the requirements of the interlaboratory comparison.

    i. The Evaluator will examine a spreadsheet of the results and discuss any discrepancies with each laboratory.

    ii. Corrective actions may include:
        a. A discussion with the laboratory of possible reasons for differences outside of acceptable criteria.
        b. Reanalysis if there is a deviation from acceptable criteria.

6. The Evaluator will complete the Laboratory Evaluation and Assistance Check List, including a brief summary of findings for each laboratory as needed. When conducting an Assistance Visit, the Evaluator must ensure that the appropriate lab personnel and QC Officer have read this and signed off before leaving the Laboratory.

7. The Evaluator will send completed Laboratory Evaluation and Assistance Check Lists to the EPA NWCA QA Assistance Visit Coordinator.

8. The EPA NWCA QA Assistance Visit Coordinator will review the Laboratory Evaluation and Assistance Check Lists, note any issues, and check off the completion of the evaluation for each participating Laboratory. All Laboratory Evaluations and completed checklists are to be sent to the NWCA Project Manager and retained permanently in an organized fashion in accordance with EPA records management policies.

7 DATA ANALYSIS PLAN

The Data Analysis Plan describes the general process used to evaluate the data for the survey. It outlines the steps taken to assess the condition of the nation's wetlands and identify the relative impact of stressors on this condition. Results from the analysis will be included in the final report and used in future analysis. This is the first analysis of wetlands of this scope and scale, so the data analysis plan will likely be refined and clarified as the data are analyzed by EPA and states.

7.1 Data Interpretation Background

The basic intent of data interpretation is to evaluate the occurrence and distribution of parameters throughout the population of wetlands in the United States within the context of regionally relevant expectations for least disturbed reference conditions. This is presented using a cumulative distribution function or similar graphic. For most indicators the analysis will also categorize the condition of the wetland as good, fair, or poor. Because of the large-scale and multijurisdictional nature of this effort, the key issues for data interpretation are unique and include: the scale of assessment, selecting the best indicators, defining the least impacted reference conditions, and determining thresholds for judging condition.

Scale of assessment. This will be the first national report on the ecological condition of the nation's wetlands using comparable methods. EPA selected the sampling locations for the survey using a probability based design, and developed rules for selection to meet certain distribution criteria, while ensuring that the design yielded a set of wetlands that would provide for statistically valid conclusions about the condition of the population of wetlands across the nation. A challenge that this mosaic of sites poses is developing a data analysis plan that allows EPA and other partners to interpret data and present results at a large, aggregate scale.

Selecting the best indicators. Indicators should be applicable across all reporting units, and must be able to differentiate a range of conditions. As part of the indicator selection process, EPA Headquarters and EPA Office of Research and Development Western Ecology Division held a conference April of 2008 to gather input from state experts. The Agency also formed a steering committee with state and regional representatives to develop and refine indicators and sampling methodologies.

EPA developed screening and evaluation criteria which included indicator applicability on a national scale, the ability of an indicator to reflect various aspects of ecological condition, and cost-effectiveness.

Defining least impacted reference condition. Reference condition data are necessary to describe expectations for biological conditions under least disturbed setting. EPA has identified and will sample 150-200 reference wetlands stratified by wetland class. EPA: (1) compiled lists of candidate reference wetlands from the 10 regions based on best professional judgment from the states and/or regions. Allocation of candidate wetlands to be sampled was based on wetland class, EPA Region, and national Ecoregion; (2) examined candidate reference wetlands for disturbances using aerial photographs in a 100 m buffer around the wetland. Disturbances were scored from 0-3 in seven categories (residential, agricultural, recreational, industrial, forestry, water development, roads). Disturbance scores for each category were summed into one "total photo" score for use as an overall disturbance index (0 = no noted disturbances); (3)

gave wetlands with a low "total photo" score, higher preference for inclusion. Wetlands were stratified by FWS Status & Trend wetland category and then states were used to spread out the sample spatially. In cases of "ties" (similar total photo scores) wetlands with agricultural and industrial disturbances (as opposed to road/recreation type disturbances) were dropped first. After that, "tie" wetlands were picked randomly to fill out cells in the table. In addition to the selection of primary reference wetlands, alternates were listed to be used in case of limited access issues with the primary wetlands. When replacing a primary wetland with an alternate, those with a similar ecoregion/wetland size were selected; (4) determined the number and types of reference wetlands appropriate and feasible for each region and selected reference wetlands for inclusion in the 20011 sampling effort.

1. *Selecting a classification system*. The U.S. FWS Wetlands Status and Trends classification system (a modified version of the Cowardin wetland classification system) will be used to determine how wetland sites are selected in the NWCA survey design. The design will stratify by state and wetland type. After field data is collected and compiled, we will test the utility of the various classification systems for use in establishing reference condition.

2. *Identifying Candidate Reference Sites*. Candidate reference sites selected for NWCA will be screened to meet regional specific criteria based on a stressor profile, surrounding land use, and physical criteria. These sites will be drawn from a population of "hand-picked sites" and from probability sample sites.

Determining thresholds for judging condition. This reference site approach is then used to set expectations and benchmarks for interpreting the data on wetland condition. The range of conditions found in the reference sites for an ecoregion describes a distribution of those biological or stressor values expected for least disturbed condition. The benchmarks used to define distinct condition classes (e.g., good, fair, poor / least disturbed, intermediate, most disturbed) are drawn from this reference distribution. EPA's approach is to examine the range of values for a biological or stressor indicator in all of the reference sites in a region, and to use the 5th percentile of the reference distribution for that indicator to separate the most disturbed of all sites from moderately disturbed sites. Using the 5th percentile means that wetlands in the most disturbed category are worse than 95% of the best sites used to define reference condition. Similarly, the 25th percentile of the reference distribution can be used to distinguish between moderately disturbed sites and those in least disturbed condition. This means that wetlands reported as least disturbed are as good as 75% of the sites used to define reference condition.

7.2 Datasets Utilized for the Report

The datasets available for use in the report were developed based on analytical methods selected during the NWCA data analysis workshop. Many of the analytical methods used in the survey stem from discussions, input, and feedback provided by the National Wetland Condition Assessment Steering Committee. Many of the methods are an outgrowth of the testing and refinement of the existing and developed methods and the logistical foundation constructed during the implementation of the Environmental Monitoring and Assessment Program (EMAP) studies from 1991 through 1994, from a Gulf Breeze pilot study conducted in 2008/9, from focused pilot studies for methods development, and from various State wetland assessment methods currently in use.

Ecological integrity. The survey will use indicators to assess ecological integrity. Ecological integrity describes the ecological condition of a wetland based on different assemblages of the vegetative community, soil characteristics, presence of appropriate hydrology and their physical habitat. The indicators include vegetation, soils, hydrology, algae, and water chemistry.

7.3 Vegetation, Soft Algae, and Diatom Data Analysis

Vegetation, Soft Algea, and Diatom data will be analyzed using both multimetric indices (MMI) and observed/expected indices (O/E) models. The MMI approach summarizes various assemblage attributes, such as composition, tolerance to disturbance, trophic and habitat preferences, as individual metrics or measures of the biological community. Candidate metrics are evaluated for aspects of performance and a subset of the best performing metrics are combined into an index known as a Vegetation, Algae, or Diatom Index of Biotic Condition (IBI). This index is then used to rank the condition of the resource.

The predictive model or O/E approach estimates the expected taxonomic composition of an assemblage in the absence of human stressors, using a set of least-disturbed sites and other variables related to natural gradients, such as elevation, wetland size, latitude and longitude. The resulting models are then used to estimate the expected taxa composition (taxa richness) at each site sampled. The number of expected taxa actually observed at a site is compared to the number of expected taxa as an Observed Expected ratio or index. Departures from a ratio of one indicate that the taxonomic composition in the sample differs from that expected under least disturbed conditions.

7.4 Soils, Hydrology and Water Quality Data Analysis

A wide array of soil and water parameters will be measured, including a mix of field and lab-derived values. Results from an analysis of soil chemistry, water chemistry, soil structure, and hydrologic alteration will feed into an assessment framework to estimate the extent of key stressors and the relative risks that stressors pose to wetland condition.

EPA will develop a set of regional stressor profiles which are qualitative characterizations of the general types of human-caused stressors that affect wetlands within a broadly defined landscape. The analytical process of grouping stressors into a profile takes into account the dominant land use and climatic conditions surrounding the surveyed population of wetlands.

We will then calculate a Human Disturbance Index (HDI) based on field observations tallying the presence and proximity of types of human activities or disturbances at Y systematically located positions with an assessment area. The HDI incorporates both the extent of human activities and the intensity of those activities. The extent will be expressed simply as the proportion of an assessment area that has at least one type of human activity recorded within its boundary. The intensity of human disturbances will be expressed by the mean proximity-weighted tally of the number of types of human land-use activities in the assessment area.

Relative Extent, Relative risk and Attributable risk evaluation

Each targeted reference site and survey site will be classified as being in either “Good”, “Fair”, or “Poor” condition, separately for each stressor variable and for each MMI (response variable). From this data, an estimate will be made of the *relative extent* (prevalence) of wetlands in Poor condition for a specified stressor and a MMI.

The *relative risk* (*RR*) of each stressor for a biological response will also be estimated. *RR* measures the severity of a stressor's effect on that response in an individual wetland assessment area, when that stressor is in Poor condition (Van Sickle, et al. 2006).

Finally, the population *attributable risk* (*AR*) of each stressor for a biological response will be estimated. *AR* combines *RR* and relative extent into a single measure of the overall impact of a stressor on a biological response, over the entire wetland resource (Van Sickle and Paulsen 2008).

7.5 Rapid Assessment Data Analysis and Methodology Evaluation

The USA Rapid Assessment Method (USA-RAM) is a field assessment method that complements the other multi-metric indices used by NWCA. It includes a coarse multi-metric index used to assess the ecological condition of wetlands. Also, a separate set of stressor metrics is organized within USA-RAM to diagnose the cause of observed degradation and opportunity for ecosystem protection, including restoration and enhancement.

USA-RAM focuses on the form and structure of wetlands. For any wetland class, we assume that a larger wetland with more complex form and structure, and less stress, tends to support higher levels of ecological integrity.[9] Individual metrics within the condition index are selected and organized to reflect a core set of hydrogeomorphic (structural) wetland attributes. Those structural attributes reflect wetland hydrology, including the source of water, hydroperiod, and connectivity to the other aquatic resources. They also reflect physical structure, including the topographic complexity in a wetland assessment area. The biological component of wetland structure is expressed in terms of the general composition, and vertical and horizontal structure of vascular plant communities. A fourth hydrogeomorphic attribute, termed landscape context, is also part of the condition index. Wetland buffer characteristics are part of the landscape attribute.

The presentation of stressor metrics within USA-RAM is based on an assessment framework that assumes wetland exposure to anthropogenic disturbance will affect both ecosystem condition and ecological resilience[2]. The magnitude of those effects is related to the proximity, intensity and duration of stressors acting on the wetland in a cumulative way. These influences and their interactions cannot be assessed with a known level of certainty using USA-RAM. Instead, USA-RAM relies on a weight-of-evidence approach to rank the causes of observed wetland degradation. The approach involves a classification and sorting of stressor types and an arithmetic "roll-up" of stressors based on their proximity, intensity and duration of effect on wetland assessment areas. Results from the tallying process are used to screen for correlations between wetland condition and likely source or sources of degradation (i.e., stressor occurrence)

USA-RAM will be calibrated for each specific wetland type using the Vegetation and Algae MMI and O/E scores described above.

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[9] Ecological Integrity: The condition of an unimpaired ecosystem as measured by combined chemical, physical (including physical habitat) and biological attributes.

[2] Ecological Resilience: The capacity of an ecosystem to withstand disturbance and human-induced stress.

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Web Page: http://www.epa.gov/nheerl/arm

9 APPENDIX A: NATIONAL WETLAND CONDITION ASSESSMENT FIELD EVALUATION AND ASSISTANCE SITE VISIT SUMMARY OF FORMS

AV-1 Presampling/General Activities
AV-2 Health and Safety
AV-3 Personnel and AA Establishment
AV-4 Veg Plot Layout/Nomenclature
AV-5 USA RAM Metrics 4-12
AV-6 Buffer and USA RAM Metrics 1-3
AV-7 Veg Characterization
AV-8 Plant Specimen Collection and Handling
AV-9 Water Quality
AV-10 Algae
AV-11 Hydrology
AV-12 Soils
AV-13 Sample Handling and Shipping
AV-14 Post Sampling Activities
AV-15 Assistance Visit Summary
T-5 NWCA Tracking - Batched Samples
Initial Site Activities
General Activities

10 APPENDIX B: WETLAND SURVEY LABORATORY LIST

Water Chemistry:

Dyanamac
200 S.W. 35th Street
Corvallis, OR 97333

Soils:
NRCS Soil Survey Research and Laboratory
National Soil Survey Center
Natural Resources Conservation Service
Federal Bldg., MS-41
100 Centennial Mall North
Lincoln, NE 68508

Algae and Vegetation Taxonomy:

EcoAnalyst
1420 S. Blaine St., Suite 14,
Moscow, ID 83843

Algal Toxin:

U.S. Geological Survey
Kansas Water Science Center
4821 Quail Crest Place
Lawrence, KS 66049

Sediment Enzyme:

National Health & Environmental Effects Laboratory
Mid-Continent Ecology Division
6201 Congdon Blvd.
Duluth, MN 55804-2595

11 APPENDIX C: Interlaboratory Total Microcystin Comparison by ELISA

11.1 Introduction:

The US EPA was tasked by Congress to acquire nationally consistent data sets so that the nation's water quality could be assessed to help guide management and regulation activities. The National Aquatic Resource Surveys were developed in response to evaluate in partnership with the states, tribes, and other federal agencies water quality of the nation's waters through nationally consistent field and laboratory methodology.

This interlaboratory investigation is necessary because more than one laboratory will be providing microcystin data for the 2011 National Wetland Assessment. The goal of this investigation is to ensure that the two labs are providing comparable data by analyzing samples in a manner that is consistent with the NWCA Laboratory Operations Manual and the manufacturer's instructions for the ELISA kits being used. The two participating laboratories are the U.S. Geological Survey's Organic Geochemistry Research Laboratory (OGRL) and the Wisconsin State Laboratory of Hygiene (WSLH). This study is defined as an interlaboratory comparison since the same protocols and method will be used by both laboratories as described in the 2011 NWCA Laboratory quality assurance project plan (QAPP).

All samples will be lysed by 3 sequential freeze/thaw cycles and filtered by 0.7 micron glass fiber filter. Filtered aliquots will then be analyzed per manufacturer directions by the Abraxis microcystin/nodularin ADDA enzyme-linked immunosorbent assay (ELISA). The WSLH will be responsible for measuring all samples collected from Wisconsin. The OGRL will measure all other samples. Both laboratories will participate in a blind interlaboratory comparison for microcystin measurements.

11.2 Interlaboratory Comparison Study Design:

1. All Wisconsin samples will be lysed and filtered at WSLH. WSLH expects a maximum of17 total samples for this study. WSLH will ship a 10 mL aliquot of filtered sample for all samples collected to Abraxis, LLC using the US EPA NWCA shipping account.

2. Abraxis, LLC will randomly select 3 samples to be analyzed as spiked samples. The spike will consist of microcystin-LR in a matrix matched diluent compared to the calibration standards used in the ELISA kit.

3. Abraxis, LLC. and U.S. EPA will assign unique sample identification numbers for each unspiked and spiked aliquot. Sample identification numbers should be randomized so that participating labs will not know the identity of samples.

4. All standard preparation, dilutions, and spikes will be conducted by Abraxis, LLC.

5. Abraxis will split the aliquots in half. One aliquot from each sample (5 mL) will remain unspiked and the other aliquot from each sample (5 mL) will be spiked with a microcystin-LR standard at a final concentration known only to Abraxis, LLC and US EPA Office of Wetlands, Oceans, and Watersheds.

6. Samples will maintained frozen at all times except with sample splitting and spiking occur or analysis.

7. 1 mL of each unspiked and spiked sample will be shipped frozen to the OGRL and WSLH for analysis. Keep remaining aliquot frozen in case issues arise.

8. Each laboratory will analyze the unspiked and spiked samples. If sample concentration exceeds 5 ppb, then dilution is necessary until the final answer is between 0.1 and 5 ppb. Quanitation will be by 4-parameter curve fit.

9. All values, including all raw data and calculated values, will be e-mailed in a spreadsheet back to the U.S. EPA NWCA QA Mangaer (Regina Poeske) and NWCA Project Lead (Michael Scozzafava).

10. Percent difference and recovery are calculated taking into consideration any sample dilutions done for the spiked samples by US EPA. Calculations should be compared to the expected and % difference between labs.

11. Final results shared back with laboratories and discussion if discrepancies arise.

11.3 Criteria for Acceptable Comparison:

1. Laboratory temperature: 20 – 25 deg. C

2. Kit blank (0 ppb) ≥ 0.8 absorbance units

3. 4-parameter curve used

4. Kit controls are ± 20% of expected value. At least 1 kit control should be analyzed after calibration standards and before samples and one at the end of samples.

5. Any diluents needed to get samples onto calibration curve should be analyzed as well. Blank diluents should be < 0.1 ppb.

6. At least 1 duplicate analyses should be analyzed for unspiked and spiked samples.

7. All samples should be within ± 20% of expected value or average value whichever is appropriate.

11.4 Corrective Action:

1. Discussion of possible reasons for differences outside of acceptable criteria.

2. Reanalysis if there is a deviation from acceptable criteria. New aliquots may need to be sent from the frozen batches made originally.

3. In case of further discrepancies, kit manufacturer may need to provide an analyses of the shipped samples to confirm results of aliquots.