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National Coastal Condition Assessment

Field Operations Manual

April 23, 2010

NOTICE

The intention of the National Coastal Condition Assessment project is to provide a comprehensive assessment for coastal waters across the United States. The complete documentation of overall project management, design, methods, and standards is contained in four companion documents:

- National Coastal Condition Assessment: *Quality Assurance Project Plan (EPA-841-R-0-004)*
- National Coastal Condition Assessment: *Site Evaluation Guidelines*
- National Coastal Condition Assessment: *Field Operations Manual (EPA-841-R-09-003)*
- National Coastal Condition Assessment: *Laboratory Methods Manual (EPA-841-R-09-002)*

This document (*Field Operations Manual*) contains a brief introduction and procedures to follow at the base location and on-site, including methods for sampling water chemistry (grabs and *in situ* measurements), benthic macroinvertebrates, sediment composition and toxicity, fish tissue, a pathogen indicator, and physical habitat. These methods are based on the guidelines developed and followed in the Coastal 2000 and National Coastal Assessment Monitoring and Assessment Program (USEPA, 2001), Methods described in this document are to be used specifically in work relating to the National Coastal Condition Assessment. All Project Cooperators must follow these guidelines. Mention of trade names or commercial products in this document does not constitute endorsement or recommendation for use. Details on specific methods for site evaluation and sample processing can be found in the appropriate companion document.

The citation for this document is:

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ACRONYMS/ABBREVIATIONS

CPR	cardiopulmonary resuscitation
DI	deionized
DO	dissolved oxygen
DVR	digital video recorder
EMAP	Environmental Monitoring and Assessment Program
EPA	Environmental Protection Agency
GED	Gulf Ecology Division, U.S. EPA Office of Research and Development
GIS	geographic information system
GL	Great Lakes
GPS	global positioning system
GRTS	Generalized Random Tessellation Stratified survey design
HDPE	high density polyethylene
MED	Mid-Continent Ecology Division, U.S. EPA Office of Research and Development
NAD	North American Datum
NAWQA	National Water-Quality Assessment Program
NCA	National Coastal Assessment
NCCA	National Coastal Condition Assessment
NEP	National Estuaries Program
NHD	National Hydrography Dataset
NIST	National Institute of Standards
NM	Nautical miles
NOAA	National Oceanographic and Atmospheric Administration
NRSA	National Rivers and Streams Assessment
ORD	Office of Research and Development, U.S. EPA
OSHA	Occupational Safety and Health Administration
PAH	Polycyclic aromatic hydrocarbon
PAR	Photosynthetically active radiation
PFD	personal floatation device
PSI	pounds per square inch
QAPP	Quality Assurance Project Plan
QA/QC	quality assurance/quality control
QCS	Quality Check Solution
SAV	Submerged aquatic vegetation
SOPs	Standard Operating Procedures
TOC	total organic carbon
TP	total phosphorus
TSS	total suspended solids
USGS	United States Geological Survey
WSA	Wadeable Streams Assessment

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1.0 NATIONAL COASTAL CONDITION ASSESSMENT BACKGROUND

The National Coastal Condition Assessment (NCCA) is one of a series of water assessments being conducted by states, tribes, the U.S. Environmental Protection Agency (EPA), and other partners. In addition to coastal waters, the National Aquatic Resource Surveys (NARS) also focuses on rivers and streams, lakes, and wetlands in a revolving sequence. The purpose of these assessments is to generate statistically-valid reports on the condition of our Nation's water resources and identify key stressors to these systems.

The goal of NARS is to address two key questions about the quality of the Nation's coastal waters:

- What percent of the Nation's coastal waters are in good, fair, and poor condition for key indicators of water quality, ecological health, and recreation?
- What is the relative importance of key stressors such as nutrients and contaminated sediments?

The NCCA is designed to be completed during the index period of June through the end of September. Field crews will collect a variety of measurements and samples from predetermined sampling locations that are determined with an assigned set of coordinates.

This manual describes field protocols and daily operations for crews to use in the NCCA. The NCCA is a probability-based survey of our Nation's coastal and estuarine waters, and is designed to:

- Assess the condition of the Nation's coastal and estuarine waters at national and regional scales, including the Great Lakes;
- Identify the relative importance of selected stressors to coastal and estuarine water quality;
- Evaluate changes in condition from previous National Coastal Assessment 's (NCA) starting in 2000; and
- Help build State and Tribal capacity for monitoring and assessment and promote collaboration across jurisdictional boundaries.

1.1 Survey Design

EPA selected sampling locations using a probability based survey design. Sample surveys have been used in a variety of fields (e.g., election polls, monthly labor estimates, forest inventory analysis) to determine the status of populations or resources of interest using a representative sample of a relatively few members or sites. Using this survey design allows data from a subset of sampled sites to be applied to the larger target population, and permits assessments with known confidence bounds.

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

- estimates of the 2010 status of all coastal waters nationally and regionally (major estuary groups and the Great Lakes); and
- estimates of the change in status in coastal waters between 2010 and 2003, nationally and regionally (i.e. major estuary groups).

With input from the states and other partners, EPA used an unequal probability design to select 682 coastal sites and 225 Great Lakes sites. Approximately 10% of the sites are selected for a return or repeat visit during the index period for quality assurance purposes.

Stratification of NCCA sites is based on major estuaries using the NOAA Coastal Assessment framework and National Estuary Program (NEP). The Great Lakes sites are stratified based on the individual Great Lake, depth zone and country. Only the shallow nearshore depth zone is included in the design for NCCA Great Lakes sites. The shallow nearshore depth zone is defined as the region extending from the shoreline to a depth of 30 m, and no more than 5 km from the shoreline.

An “oversample” of additional sites also is available so that any state wishing to conduct a state level or NEP-level design could be accommodated and to provide alternate sampling sites if specific sites are rejected. Sites were also identified for the Canadian nearshore although sampling of these sites is not a part of the NCCA.

1.1.1 Target Population and Sample Frame

The target population for the marine coasts consists of all coastal waters of the conterminous United States from the head-of-salt to confluence with the ocean, including inland waterways and major embayments such as Florida Bay and Cape Cod Bay. For the purposes of this study the head of salt is defined as < .05 parts per thousand (ppt) salinity. The target population for the Great Lakes consists of all waters of the Great Lakes of the United States and Canada. The current target population is restricted to the shallow nearshore zones of Lake Superior, Lake Michigan, Lake Huron, Lake Erie, and Lake Ontario. The NCCA Great Lakes sites are restricted to the United States portions. Please refer to the Site Evaluation Guidelines and the NCCA Web site (http://www.epa.gov/owow/monitoring/nationalsurveys.html) for more detailed information on the target population.

The sample frame was derived from prior National Coastal Assessments developed by EPA Office of Research and Development (ORD) Gulf Ecology Division (GED). The prior GED sample frame was enhanced as part of the National Coastal Monitoring Network design by including information from NOAA’s Coastal Assessment Framework, boundaries of National Estuary Programs and identification of major coastal systems. For NCCA 2010 information on salinity zones was obtained from NOAA. For Delaware Bay, Chesapeake Bay, Puget Sound and state of South Carolina, the prior NCA sample frames were replaced by GIS layers provided by those organizations, ensuring that prior areas sampled in NCA were not excluded and any differences from the previous sample frames to the current sample frame are clearly identified in this 2010 NCCA sample frame. The sample frame for the Great Lakes sites were obtained from EPA ORD Mid-Continent Ecology Division (MED).

1.1.2 Replacing Sites

It is likely that some sites will be determined to be unsampleable, therefore, a number of backup sites are provided in the form of an oversample list that is provided to each state. A site can be deemed unsampleable for any number of reasons, including being too shallow to properly operate sampling equipment, in the middle of a navigational channel where it is unsafe, or practically on top of a neighboring site. When a site is determined to be unsampleable, please document the sampling status of the site and select the next backup station on the list for that state and estuary type. This maintains the probabilistic integrity of the survey. Please refer to the *Site Evaluation Guidelines* for more detailed information on determining site sampling status.

If a site is generally sampleable, but one or more indicators cannot be collected (e.g. no fish caught or site is too deep to collect sediment), the site should not be dropped. Rather, the crew will flag that indicator and document the reason why the indicator could not be collected. See Section 4.1.6 for information regarding the collection of sediment samples.

1.2 Selection of NCCA Indicators

Indicators for the 2010 survey will basically remain the same as those used in the historic National Coastal Condition Report with a few modifications. The most prominent change in this year's survey is the inclusion of coasts along the Great Lakes. Therefore both sample collection methods and laboratory methods will reflect freshwater and saltwater matrices.

The NCCA workgroup decided on a few changes to the original indicators based on recommendations from a state and tribal workshop held in 2008 and other discussions. The changes are: 1) *Enterococcus* will be collected as a human health indicator; 2) for sediment toxicity testing, lab methods will use *Leptochirus* instead of *Ampelisca sp*. for saline sites and *Hyalella* for freshwater sites; 3) tissue studies will be conducted using whole fish, and 4) the NCCA will not include the collections of samples for fish community structure, Total Suspended Solids (TSS), or PAHs in fish tissue.

1.3 Description of NCCA Indicators

In Situ Water Quality Measurements

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

Light Attenuation

A Photosynthetically Active Radiation (PAR) meter, will be used to obtain a vertical profile of light in order to calculate the light attenuation coefficient at each station. PAR measurements are taken at the same depths as other water column indicators.

Secchi Disk Transparency

A Secchi disk is a commonly used black and white patterned disk used to measure the clarity of water within a visible distance.

Water Chemistry and Associated Measurements

Water chemistry measurements will be used to determine nutrient enrichment, as well as classification of trophic status. Parameters measured include total and dissolved nitrogen and phosphorus.

Chlorophyll-a

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

Dissolved Nutrients

A portion of the filtrate produced from the processing of the chlorophyll-*a* sample will be collected in the field and processed in the laboratory for dissolved nutrients.

Phytoplankton Assemblage

Phytoplankton are plant microorganisms that float in the water, such as certain algae, and are the primary source of energy in most lake systems (Schriver et al. 1995). Phytoplankton are highly sensitive to changes in ecosystems (e.g., turbidity and nutrient enrichment). Phytoplankton will be collected in Great Lakes sites only.

Underwater Video

At Great Lakes sites only, crews will use an underwater video camera with recorder to capture 1 minute of video focused on the substrate at the X-site. Video will be used in the lab to visually document the bottom composition, and record the presence or absence of zebra mussels, Cladophora, or other organisms.

Sediment Assessment

Sediment grab samples will be obtained to measure sediment composition (e.g., grain size, percent moisture, organic content, etc.), toxicity and chemistry in order to determine sediment condition.

Benthic Macroinvertebrate Assemblage

Benthic macroinvertebrates are bottom-dwelling animals without backbones (“invertebrates”) that are large enough to be seen with the naked eye (“macro”). Examples of macroinvertebrates include: aquatic worms, mollusks, and crustaceans. Populations in the benthic assemblage respond to a wide array of stressors in different ways so that it is often possible to determine the type of stress that has affected a macroinvertebrate assemblage (Klemm et al., 1990). Because many macroinvertebrates have relatively long life cycles of a year or more and are relatively immobile, the structure of the macroinvertebrate assemblage is a response to exposure of present and/or past conditions. The benthic macroinvertebrate data will serve as the basis for assessing aquatic community health.

Habitat Assessment

The habitat assessment of the site provides information about the type of habitat located at a site. This assessment includes presence/absence of submerged aquatic vegetation, marine debris, and the basic habitat type (e.g., open water, tidal flat, marina, harbor, inlet, tidal river/stream, seagrass bed, sandy/muddy bottom, rocky bottom, shelly bottom, coral reef, etc.)

Fecal Indicator (Enterococci)

Enterococci are bacteria that are endemic to the guts of warm blooded creatures. These bacteria, by themselves, are not considered harmful to humans but often occur in the presence of potential human pathogens (the definition of an indicator organism). Epidemiological studies of marine and fresh water bathing beaches have established a direct relationship between the density of *Enterococci* in water and the occurrence of swimming-associated gastroenteritis.

Fish Tissue

The fish tissue indicator, which measures bioaccumulation of persistent toxics, is used to estimate the ecological risks associated with fish consumption by wildlife. In this study, fish will be collected and whole body tissue will be homogenized and analyzed to estimate concentrations of target contaminants. Various studies have been conducted on contaminants in different tissues of the fish (e.g., whole fish, fillets, or livers). For this study, the focus will be on analyzing whole fish for contaminants to generate data for ecological purposes.

In the Great Lakes only, additional fish composite samples will be collected at 150 of the 225 sites (ideally the first 30 sites per lake). Fillet tissue from these samples will be homogenized and analyzed to generate fish contamination data related to human health.

1.4 Supplemental Material to the Field Operations Manual

The Field Operations Manual describes field protocols and daily operations for crews to use in the NCCA. Following these detailed protocols will ensure consistency across regions and reproducibility for future assessments. Before beginning sampling at a site, crews should prepare a packet for each site containing pertinent information to successfully conduct sampling. This includes a road map or navigation chart and a set of directions to the site, topographic/bathymetric maps, land owner access forms (where applicable), sampling permits (if needed), site evaluation forms and other information necessary to ensure an efficient and safe sampling day.

Field crews will also receive a quick-reference handbook that contains tables and figures summarizing field activities and protocols from the Field Operations Manual. This waterproof handbook will be the primary field reference used by field teams after completing the required field training session. The field teams are also required to keep the field operations manual available in the field for reference and for possible protocol clarification, as well as other equipment manuals (probes, etc.).

Large-scale and/or long-term monitoring programs such as those envisioned for national surveys and assessments require a rigorous Quality Assurance (QA) program that can be implemented consistently by all participants throughout the duration of the monitoring period. Quality assurance is a required element of all EPA-sponsored studies that involve the collection of environmental data (USEPA 2000a, 2000b). Field teams will be provided a copy of the

integrated Quality Assurance Project Plan (QAPP). The QAPP contains more detailed information regarding QA/QC activities and procedures associated with general field operations, sample collection, measurement data collection for specific indicators, data reporting activities, and the information management plan for this project. For more information on the Quality Assurance procedures, refer to the *National Coastal Condition Assessment: Quality Assurance Project Plan (EPA 841-R-09-004)*

Related NCCA documents include the following: 1) National Coastal Condition Assessment: Quality Assurance Project Plan (EPA 841-R-09-004); 2) National Coastal Condition Assessment: Site Evaluation Guidelines; and 4) National Coastal Condition Assessment: Laboratory Methods Manual (EPA 841-R-09-002). These documents are available at: http://www.epa.gov/owow/monitoring/nationalsurveys.html

2.0 DAILY OPERATIONS SUMMARY

This Field Operations Manual will be used for sampling at both marine and coastal freshwater Great Lakes sites. For the most part, the same indicators will be collected, but some of the sampling will be conducted with different equipment. Additional parameters to be collected at all Great Lakes sites will include phytoplankton and underwater video of the substrate. Human health fish tissue samples will be collected at a subset of Great Lakes sites. This section presents a general overview of the activities that a field team is to conduct during a typical 1-day sampling visit to a site. General guidelines for recording data using standardized field data forms and sample labels are also presented. Finally, general safety and health considerations and guidelines related to field operations are described. Please be sure to fully know and follow the safety considerations applicable to your organization.

2.1 Sampling Scenario

The field methods for the NCCA are designed to be completed in one field day. Depending on the time needed for both the sampling and travel for the day, an additional day may be needed to complete sampling or for pre-departure and post-sampling activities (e.g., cleaning equipment, repairing gear, shipping samples, and traveling to the next site). Remote sites with lengthy or difficult approaches may require more time, and field crews will need to plan accordingly. Conversely, some sites may be in relatively close proximity, allowing multiple sites to be sampled in a single day.

A field crew will typically consist of three to four people. Each field team should define roles and responsibilities for each team member to organize field activities efficiently. A minimum of two people are always required in a boat to execute the sampling activities and to ensure safety. One crew member is primarily responsible for boat operation and navigation. Any additional members may assist with the collection of samples or provide logistical support. Daily field activities may differ depending on whether the team is collecting fish with the use of active (trawling, seining, hook and line, etc.) or passive (gill net, hoop net, long-lines, etc.) fish collection methods. Likewise, teams may choose to collect the fish tissue sample on a visit separate from the collection of water quality samples, sediment and other associated parameters. Other minor modifications to the sampling scenario may be made by teams; however the sequence of sampling events presented in Figures 2-1, 2-2 and 2-3 (depending on the type and timing of fish collection) should be adhered to and is based on the need to protect some types of samples from potential contamination and to minimize holding times once samples are collected**.**

Since Enterococcus levels are shown to be highest in the morning before high levels of solar irradiation, it is recommended that these samples be collected as early in the day as possible and with minimal disturbance of water and sediment, **as long as they are filtered prior to six hour hold time threshold.** Field team should choose the sampling scenario that best fits with these goals.

The field team arrives at the site in the early morning to complete the sampling in a single day. Crew members responsible for collecting water chemistry, sediment grabs and fish tissue must remember to not apply sunscreen or other chemical contaminants until after the sample is collected, to avoid compromising the integrity of the sample. The sampling sequence is to:

- Verify site as correct location to obtain samples (whole crew);
- Make notations of weather, habitat type, presence of submerged aquatic vegetation (SAV), macroalgae, and debris;
- Set net for fish collection (if using passive sampling gear);
- Take Secchi disk transparency depth measurements;
- Conduct *in situ* measurements of dissolved oxygen, pH, temperature, and salinity/conductivity;
- Take light attenuation measurements with Photosynthetically Active Radiation (PAR) meter;
- Collect fecal indicator (Enterococci) sample;
- Collect water for chemistry and chlorophyll-*a;*
- Collect benthic samples;
- Collect sediment samples for chemistry, toxicity, and grain size;
- Collect fish tissue samples (note above that if passive sampling gear is used, e.g., a stationary net, it can be set upon arrival on site);
- Filter Enterococci and chlorophyll-*a* samples;
- Collect dissolved nutrients sample (chlorophyll-*a* filtrate);
- Preserve and prepare all samples for shipment;
- Review field forms;
- Report sampling event; and
- Ship time-sensitive samples.

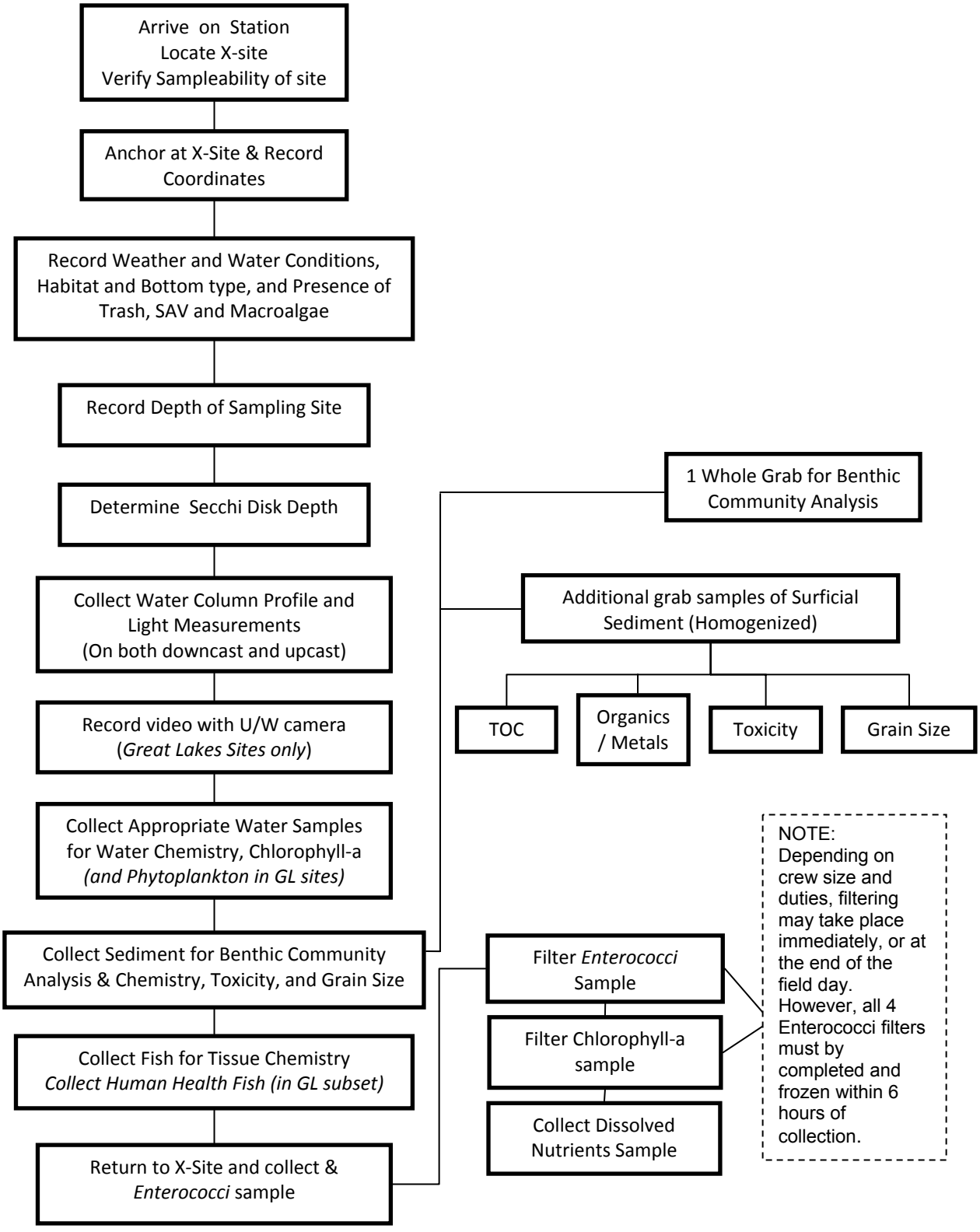


Figure 2-1. Field Sampling Scenario for Teams Using Active Fishing Methods

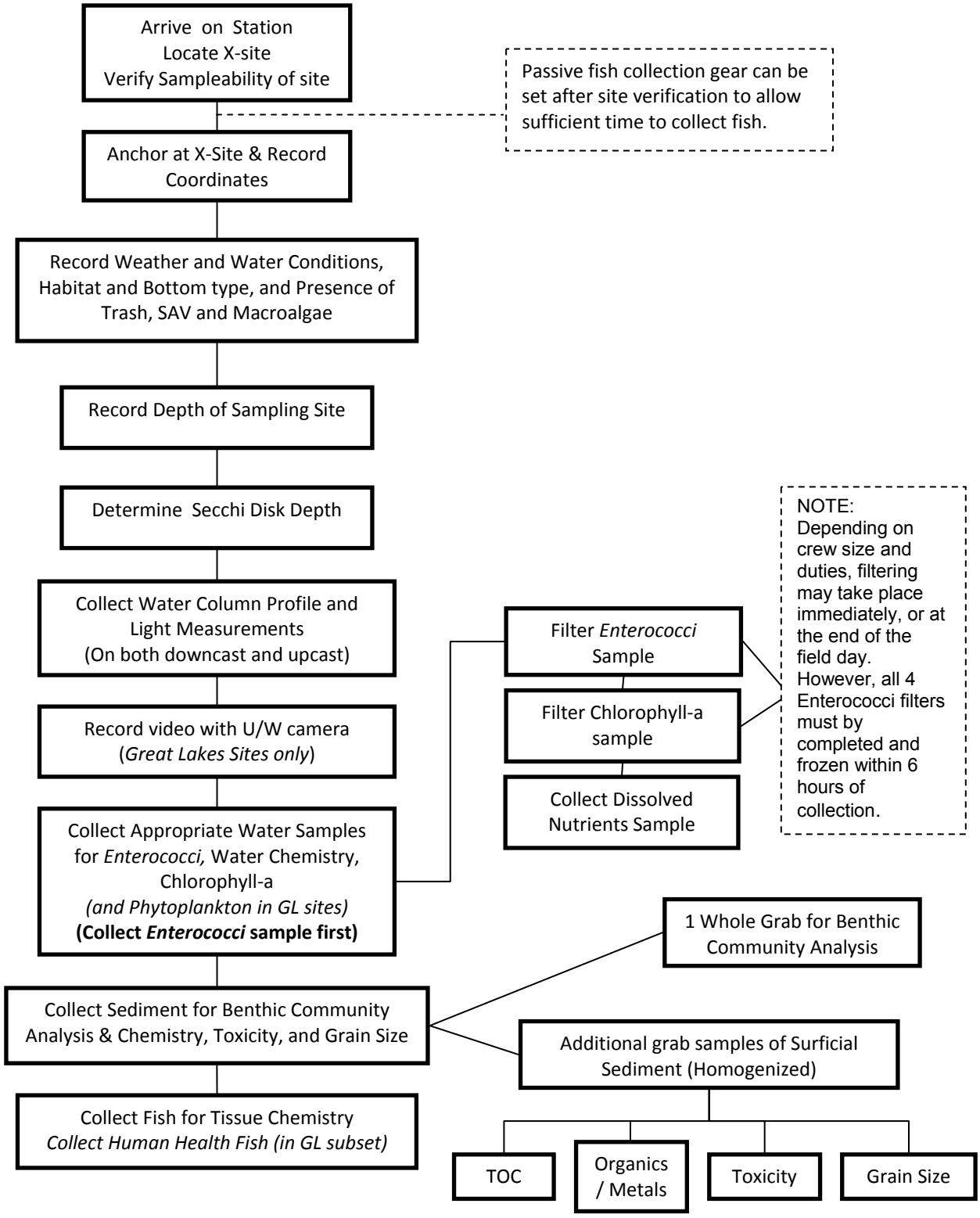


Figure 2-2. Field Sampling Scenario for Teams Using Passive Fishing Methods

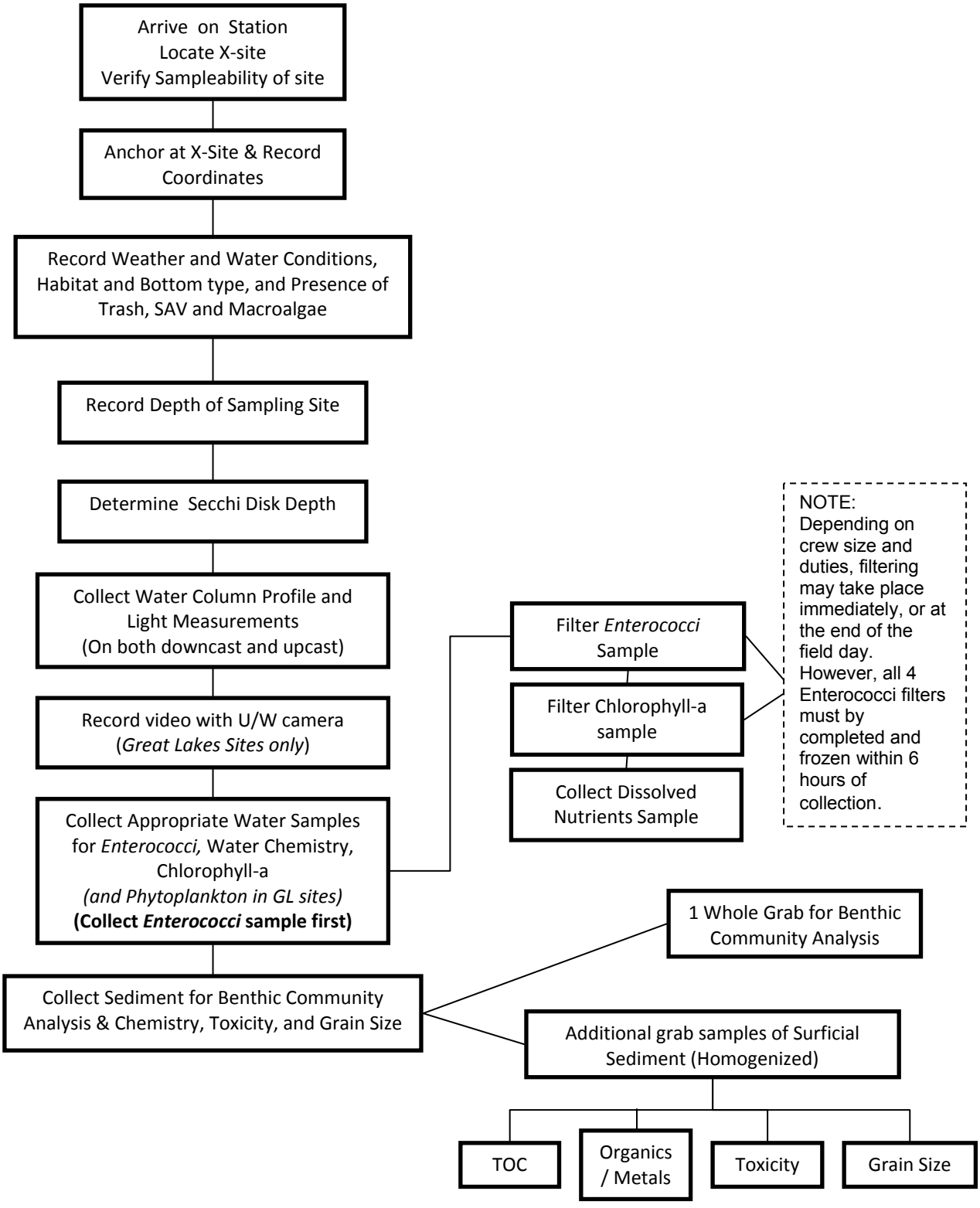


Figure 2-3. Field Sampling Scenario for Teams not Collecting Fish for Tissue Chemistry

2.2 Recording Data and Other Information

All samples need to be identified and tracked, and associated information for each sample must be recorded. To assist with sample identification and tracking, labels are preprinted and provided by EPA with sample ID numbers (Figure 2-4).

WATER CHEMISTRY - NOT FILTERED
NCCA10- __ __ __ __
_____ /_____ /2010
999101

CHLOROPHYLL-a
NCCA10- __ __ __ __
_____ /_____ /2010
Volume Filtered: _____mL
999102

NUTRIENTS - FILTERED
NCCA10- __ __ __ __
_____ /_____ /2010
Salinity: _____‰
999103

BENTHOS
NCCA10- __ __ __ __
_____ /_____ /2010
Jar 1 of _____
999104

SEDIMENT ORGANICS (SEDO)
NCCA10- __ __ __ __
_____ /_____ /2010
999105

SEDIMENT GRAIN SIZE (SEDG)
NCCA10- __ __ __ __
_____ /_____ /2010
999106

SEDIMENT TOC (SEDC)
NCCA10- __ __ __ __
_____ /_____ /2010
999107

SEDIMENT TOXICITY (SEDX)
NCCA10- __ __ __ __
_____ /_____ /2010
999108

ECO FISH TISSUE - OUTER BAG
NCCA10- __ __ __ __
_____ /_____ /2010
Genus Species: _____
Length (mm) Min.: _____ Max.: _____
999109

ECO FISH TISSUE - INNER BAG ___ OF ___
NCCA10- __ __ __ __
_____ /_____ /2010
Genus Species: _____
Length (mm) Min.: _____ Max.: _____
999109

ECO FISH TISSUE - INNER BAG ___ OF ___
NCCA10- __ __ __ __
_____ /_____ /2010
Genus Species: _____
Length (mm) Min.: _____ Max.: _____
999109

ECO FISH TISSUE - INNER BAG ___ OF ___
NCCA10- __ __ __ __
_____ /_____ /2010
Genus Species: _____
Length (mm) Min.: _____ Max.: _____
999109

HUMAN HEALTH FISH TISSUE
NCCAGL10-__ __ __ __
_____ /_____ /2010
Genus Species: _____
Length (mm): _____
222001.01

PHYTOPLANKTON
NCCAGL10- __ __ __ __
_____ /_____ /2010
999100

Figure 2-4. Example Sample Labels for Sample Tracking and Identification.

It is imperative that field and sample information be **recorded accurately, consistently, and legibly**. The cost of a sampling visit coupled with the short index period severely limits the ability to resample a site if the initial information recorded was inaccurate or illegible. Illegible or incorrect information can result in substantially increased time to transfer information from field forms to the National Aquatic Resource Surveys Surface Water Information Management System. Guidelines for recording field measurements are presented in Table 2-1.

Table 2-1. Guidelines for Recording Field Measurements and Tracking Information

Activity	Guidelines
Field Measurements	
Data Recording	Record measurement values and observations on data forms preprinted on water-resistant paper. Use No. 2 pencil only (fine-point indelible markers can be used if necessary) to record information on forms. Record data and information using correct format as provided on data forms. Be sure to accurately record site IDs and sample numbers. Print legibly (and as large as possible). Clearly distinguish letters from numbers (e.g., 0 versus O, 2 versus Z, 7 versus T or F, etc.), but do not use slashes. When recording comments, print or write **legibly**. Make notations in comments field only; avoid marginal notes. Be concise, but avoid using abbreviations or "shorthand" notations. If you run out of space, attach a sheet of paper with the additional information, rather than trying to squeeze everything into the space provided on the form.
Data Qualifiers (Flags)	Use only defined flag codes and record on data form in appropriate field. K = Measurement not attempted or not recorded. Q = Failed quality control check; remeasurement not possible. U = Suspect measurement; remeasurement not possible. F*n* = Miscellaneous flags (*n* = 1, 2, etc.) assigned by a field team during a particular sampling visit (also used for qualifying samples). Explain reason for using each flag in comments section on data form.
Sample Labels	Use adhesive labels with preprinted ID numbers and follow the standard recording format for each type of sample. Use a marker to record information on label. Cover the completed label with clear tape. Record sample ID number from label and associated collection information on sample collection form preprinted on water-resistant paper.

Activity	Guidelines
Sample Collection and Tracking	
Sample Qualifiers (Flags)	Use only defined flag codes and record on sample collection form in appropriate field. K = Sample not collected or lost before shipment; resampling not possible. U = Suspect sample (e.g., possible contamination, does not meet minimum acceptability requirements, or collected by non-standard procedure). F*n* = Miscellaneous flags (*n*=1, 2, etc.) assigned by a field team during a particular sampling visit (also used for field measurements). Explain reason for using flags in "Comments" on sample collection form.
Review of Labels and Data Collection Forms	Compare information recorded on labels and sample collection form for accuracy before leaving site. Review labels and data collection forms for accuracy, completeness, and legibility before leaving site. The Field Team Leader must review all labels and data collection forms for consistency, correctness, and legibility before transfer to the Information Management Center.

2.3 Safety and Health

Collection and analysis of samples can involve significant risks to personal safety and health. This section describes recommended training, communications, safety considerations, safety equipment and facilities, and safety guidelines for field operations.

2.3.1 General Considerations

Important considerations related to field safety are presented in Table 2-2. It is the responsibility of the crew leader to ensure that the necessary safety courses are taken by all field personnel and that all safety policies and procedures are followed. Please follow your own agency's health and safety protocols, or refer to the *Health and Safety Guidance for Field Sampling: National Coastal Condition Assessment* (available from the EPA Regional Coordinator). Additional sources of information regarding safety-related training include the American Red Cross (2006), the National Institute for Occupational Safety and Health (1981), and U.S. Coast Guard (1989).

Field crew members should become familiar with the hazards involved with sampling equipment and establish appropriate safety practices prior to using them. Make sure all equipment is in safe working condition. Personnel must consider and prepare for hazards associated with the operation of motor vehicles, boats, winches, tools, and other incidental equipment. Boat operators should meet any state requirements for boat operation and be familiar with U.S. Coast Guard rules and regulations for safe boating contained in a pamphlet, "*Federal Requirements for Recreational Boats*," available from a local U.S. Coast Guard Director or Auxiliary or State Boating Official (U.S. Coast Guard, 1989). Personal Flotation Devices (PFD) must be worn by crew members at all times on the water. All boats with motors must have fire extinguishers, boat horns, PFDs, and flares or communication devices.

Table 2-2. General Health and Safety Considerations.

Recommended Training
▪ First aid and cardiopulmonary resuscitation (CPR)
▪ Vehicle safety (e.g., operation of 4-wheel drive vehicles, trailering boats, etc.)
▪ Field safety (weather, personal safety, navigation, site reconnaissance prior to sampling)
▪ Equipment design, operation, and maintenance
▪ Handling of chemicals and other hazardous materials
Communications
▪ Check-in schedule
▪ Sampling itinerary (vehicle used & description, time of departure & return, travel route)
▪ Contacts for police, ambulance, hospitals, fire departments, search and rescue personnel
▪ Emergency services available near each sampling site and base location
▪ Cell (or satellite) phone and VHF radio.
Personal Safety
▪ Field clothing and other protective gear including PFDs for all team members
▪ Medical and personal information (allergies, personal health conditions)
▪ Personal contacts (family, telephone numbers, etc.)
▪ Physical exams and immunizations

A communications plan to address safety and emergency situations is essential. All field personnel need to be fully aware of all lines of communication. Field personnel should have a daily check-in procedure for safety. An emergency communications plan should include contacts for police, ambulance, fire departments, hospitals, and search and rescue personnel.

Proper field clothing should be worn to prevent hypothermia, heat exhaustion, sunstroke, drowning, or other dangers. Field personnel must be able to swim, and personal flotation devices must be used. A personal flotation device (PFD) and suitable footwear must be worn at all times while on board a boat.

Many hazards lie out of sight in the bottoms of coastal areas. Broken glass or sharp pieces of metal embedded in the substrate can cause serious injury if care is not exercised when walking or working with the hands in such environments. Infectious agents and toxic substances that can be absorbed through the skin or inhaled may also be present in the water or sediment. Personnel who may be exposed to water known or suspected to contain human or animal wastes that carry causative agents or pathogens must be immunized against tetanus, hepatitis, typhoid fever, and polio. Biological wastes can also be a threat in the form of viruses, bacteria, rickettsia, fungi, or parasites.

2.3.2 Safety Equipment

Appropriate safety apparel such as foul weather gear, safety glasses, etc. must be available and used when necessary. First aid kits, fire extinguishers, and blankets must be readily available in the field. Cellular or satellite telephones and/or portable radios should be provided to field teams working in remote areas in case of an emergency. Supplies (e.g., clean water, anti-bacterial soap, ethyl alcohol) must be available for cleaning exposed body parts that may have been contaminated by pollutants in the water.

2.3.3 Safety Guidelines for Field Operations

General safety guidelines for field operations are presented in Table 2-3. Personnel participating in field activities should be in sound physical condition and have a physical examination annually or in accordance with organizational requirements. All surface waters and sediments should be considered potential health hazards due to potential toxic substances or pathogens. Persons must become familiar with the health hazards associated with using chemical fixing and/or preserving agents. Chemical wastes can be hazardous due to flammability, explosiveness, toxicity, causticity, or chemical reactivity. All chemical wastes must be discarded according to standardized health and hazards procedures (e.g., National Institute for Occupational Safety and Health [1981]; U.S. EPA [1986]).

During the course of field research activities, field teams may observe violations of environmental regulations, may discover improperly disposed hazardous materials, or may observe or be involved with an accidental spill or release of hazardous materials. In such cases it is important that the proper actions be taken and that field personnel do not expose themselves to something harmful. The following guidelines should be applied:

First and foremost, protect the health and safety of all personnel. Take necessary steps to avoid injury or exposure to hazardous materials. If you have been trained to take action such as cleaning up a minor fuel spill during fueling of a boat, do it. However, you should always err on the side of personal safety.

Field personnel should never disturb or retrieve improperly disposed hazardous materials from the field to bring back to a facility for “disposal”. To do so may worsen the impact, incur personal liability for the team members and/or their respective organizations, cause personal injury, or cause unbudgeted expenditure of time and money for proper treatment and disposal of the material. Notify the appropriate authorities so they may properly respond to the incident.

For most environmental incidents, the following emergency telephone numbers should be provided to all field teams: State or Tribal department of environmental quality or protection, U.S. Coast Guard, and the U.S. EPA regional office. In the event of a major environmental incident, the National Response Center may need to be notified at 1-800-424-8802.

Table 2-3. General Safety Guidelines for Field Operations

- At least two crew members must be present during all sample collection activities, and no one should be left alone while out on the water.
- Use caution and wear a suitable PFD.
- Use caution using the Ponar-type samplers. The jaws are sharp and may close unexpectedly.
- Exposure to water and sediments should be minimized as much as possible. Use gloves if necessary, and clean exposed body parts as soon as possible after contact.
- All electrical equipment must bear the approval seal of Underwriters Laboratories and must be properly grounded to protect against electric shock.
- Use appropriate protective equipment (e.g. gloves, safety glasses) when handling and using hazardous chemicals.
- Crews working in areas with poisonous snakes must check with the local Drug and Poison Control Center for recommendations on what should be done in case of a bite from a poisonous snake.
- Any person allergic to bee stings, other insect bites, or plants (i.e., poison ivy, oak, sumac, etc.) must take proper precautions and have any needed medications handy.
- Field personnel should be familiar with the symptoms of hypothermia and know what to do in case symptoms occur. Hypothermia can kill a person at temperatures much above freezing (up to 10^{o}C or 50^{o}F) if he or she is exposed to wind or becomes wet. Immersion in the cool waters experienced during the summer along most of the coast and Great Lakes can also rapidly result in hypothermia.
- Field personnel should be familiar with the symptoms of heat/sun stroke and be prepared to move a suffering individual into cooler surroundings and hydrate immediately.
- Handle and dispose of chemical wastes properly. Do not dispose of any chemicals in the field.

3.0 BASE SITE ACTIVITIES

Field teams conduct a number of activities at their base site (i.e. office or laboratory, camping site, or motel). These include tasks that must be completed both before departure to the site and after return from the field (Figure 3-1). Close attention to these activities is required to ensure that the field teams know: 1) where they are going, 2) that access is permissible and possible; 3) that equipment and supplies are available and in good working order to complete the sampling effort; and 4) that samples are packed and shipped appropriately.

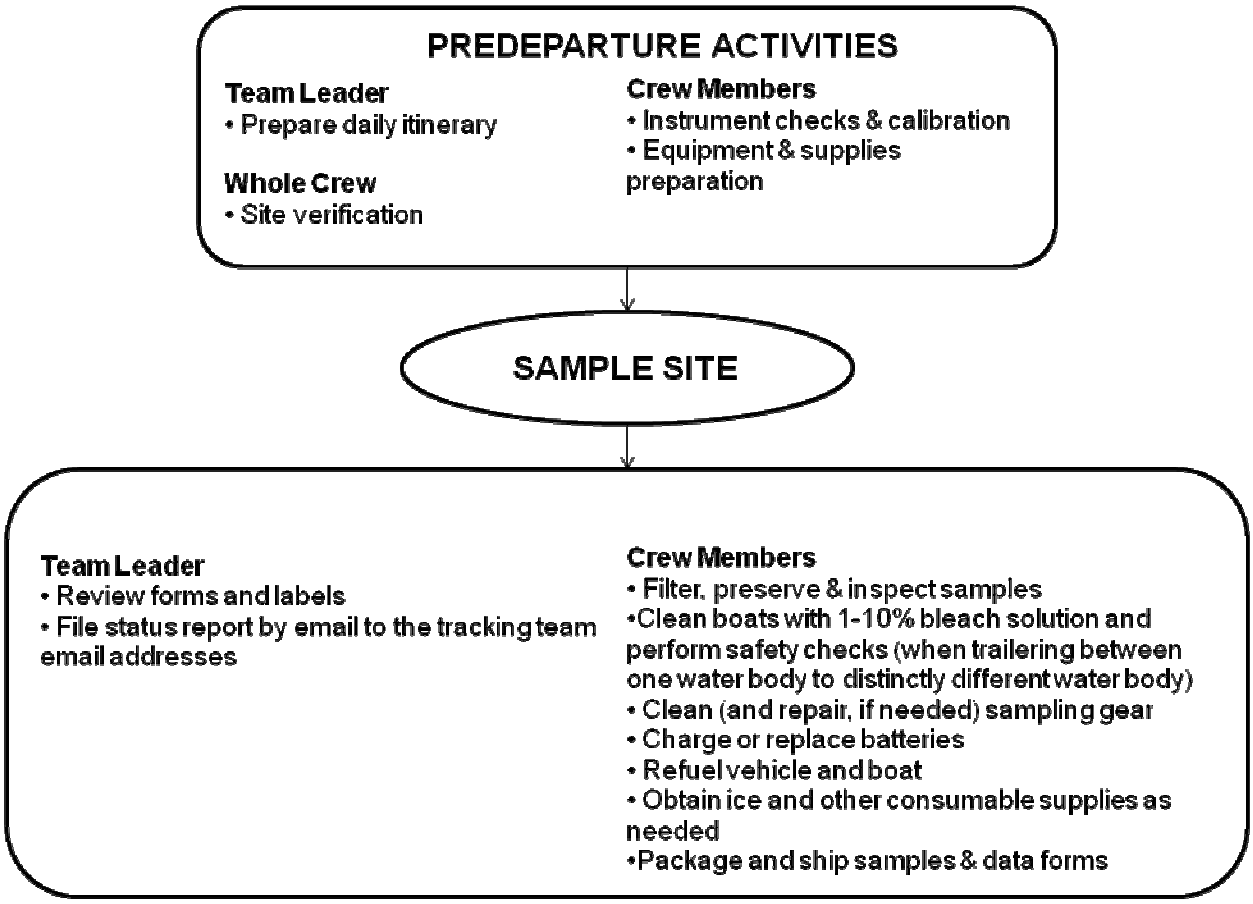


Figure 3-1. Overview of Base Site Activities.

3.1 Predeparture Activities

Predeparture activities include the development of a daily itinerary, instrument checks and calibration, and equipment and supply preparation. Procedures for these activities are described in the following sections.

3.1.1 Daily Itineraries

The Field Team Leaders are responsible for developing daily itineraries. This entails compiling maps, navigational charts, contact information, copies of permission letters, permits, access instructions, location of FedEx offices, and location of hospitals or other emergency services (a "site packet"). Additional activities include confirming the best access routes, calling the landowners or local contacts, confirming lodging plans, and coordinating rendezvous locations with individuals who must meet with field teams prior to accessing a site. Also, the Leader should identify appropriate boat ramps or marinas, and gas docks. If the crew is planning a multiple day/multiple site trip, information for each day and site must be developed.

Field Team Leaders should call the Site Kit Coordinator or Field Logistics Coordinator at least two weeks in advance to request site kits based on the upcoming schedule. Changes in the itinerary during the week, such as canceling a sampling day, must be relayed by the crew leader to the Field Logistics Coordinator as soon as possible.

3.1.2 Instrument Checks and Calibration

Each field team must test and calibrate instruments prior to sampling. Calibration can be conducted prior to departure for the site or at the site, with the exception of dissolved oxygen (DO) calibration. DO meters should be calibrated at the site. Field instruments include a global positioning system (GPS) receiver, a Photosynthetically Active Radiation (PAR) meter, and a multiprobe unit for measuring DO, pH, temperature, salinity (marine sites) and conductivity (freshwater sites). Field teams should have access to backup instruments if any instruments fail the manufacturer performance tests or calibrations. Prior to departure, field teams must:

- Turn on the GPS receiver and check the batteries if using hand-held GPS unit. Replace batteries immediately if a battery warning is displayed. Otherwise, the GPS is installed on the boat and runs off of the boat electrical system.
- Test and calibrate the multi-probe meter. Each field team should have a copy of the manufacturer's calibration and maintenance procedures. All meters should be calibrated according to manufacturer specifications provided along with the meter. Once a week, crews should check their multiprobe against the provided Quality Check Solution. This QCS is provided to all crews in their base kits and is used to check pH and conductivity measurements.
- Ensure that the PAR meter's handheld display unit has fresh batteries, that the unit is functioning properly, and that the correct calibration factors are entered for each probe. These factors are supplied by the manufacturer and are specific to each individual probe. PAR sensors require no field calibration, however, they should be returned to the manufacturer at least every 2 years for calibration. Procedures for the initial setup of the LI-COR LI-1400 Datalogger is presented in Table 3-1. These steps can be used to verify the setup of the unit, or to enter coefficient values should a new sensor need to be installed.

- For teams operating in the Great Lakes, ensure that the underwater video system's battery is charged and all components are hooked up correctly. Ensure that the attached GPS is set up to output information to the GPS overlay (this will be done prior to shipment of the system to field crews, but should a crew need to verify setup, refer to Section 5.5.3).

Table 3-1. Initial Setup Procedures for LI-COR LI-1400 Datalogger

LI-COR systems received from GLEC will have been configured for use already and will be ready for sampling, these instructions will be for future reference.

The following example demonstrates how you can configure the LI-1400 with the instrument keypad to view or log instantaneous data from a single LI-190SA Quantum Sensor.

Example 1a. Configure channel I1 for a LI-COR LI-190SA Quantum Sensor whose calibration multiplier is -125.0µmols-1m-2/µAmp (Each sensor has a unique multiplier value supplied from the factory)

1. Connect the Quantum LI-190 sensor to the BNC connector on top of the LI-1400 labeled I1.
2. Turn on the LI-1400 meter.
3. Press the Setup key.
4. Use the left or right arrow keys to navigate to "SETUP CHANNELS".
5. Press the Enter key to begin the sensor setup.
6. Use the left or right arrow keys to navigate to "I1=Light", press Enter".
7. Using the Shift key and the number/ letter keys, type a description for this channel.
This description can be anything such as QUANTUM to describe the type of sensor, or AMB to describe what the reading will be used for in the NCCA sampling.
8. Press the down arrow key to enter the multiplier.
Use the Multiplier value found on the Certificate of Calibration that came with your sensors. There should be a unique certificate and calibration multiplier value for each sensor.
9. Press the down arrow key; enter the unit label. Type UM for the label.
10. Press the down arrow key; select "1 sec" to have instantaneous values displayed.
The running average parameter will not be used, but could be set here to any desired value.
11. Press the down arrow key; select "Log Routin=none"
12. The remaining options do not need to be set as they apply only when using a Log Routine.
13. Repeat this entire procedure for "I2=Light" to setup the underwater sensor.

3.1.3 Equipment and Supply Preparation

Field teams must check the inventory of supplies and equipment prior to departure using the equipment and supplies checklists provided in Appendix A; use of the lists is mandatory. Obtain sufficient wet and dry ice for sample preservation and storage. Pack meters, probes, and sampling gear in such a way as to minimize physical shock and vibration during transport. Pack stock solutions as described in Table 3-2. Follow the regulations of the Occupational Safety and Health Administration (OSHA).

Table 3-2. Stock Solutions, Uses, and Methods for Preparation.

Solution	Use	Preparation
Bleach (1-10%)	Clean nets, gear, and inside of boat	Add 10 - 100 mL bleach to 1 L distilled water.
Quality Check Solution for multi-probe meter	QCS for pH and conductivity calibration	None (included in base kits)
Buffered Formalin	Preserve benthic samples	Add ¼ teaspoon Rose Bengal crystals and 8 tablespoons Borax to 2 gallons 100% Formalin solution
Lugol's Solution	Preserve phytoplankton samples (Great Lakes sites only)	None (included in GL base kits)

Site kits of consumable supplies for each sampling site will be delivered based on the schedule each crew provides prior to the sampling season. **Field crew leaders MUST provide a schedule in order to receive the site kits and must call the Field Logistics Coordinator two weeks prior to sampling.** Field crew leaders involved with Great Lakes human health fish sampling must specifically request a human health fish tissue supply kit for those sites from the Great Lakes Human Health Fish Tissue Field Kit Coordinator. Please note, field kits for all sites cannot be provided at the beginning of the field season but will be sent out as requested throughout the index period. If your schedule changes, report the change as soon as possible to the Field Logistics Coordinator. The site kit will include data forms, labels, sample jars, bottles and other supplies (see complete list in Appendix A). The teams must inventory these site kits before departure to ensure that all supplies are included. If any items are missing or incorrect, crews must request these supplies from the Site Kit Coordinator as soon as possible so that the supplies are received prior to departure. Container labels should not be covered with clear tape until all information is completed during sampling at the site. Store extra sampling site kits in the vehicles in case of loss, damage, etc. Inventory these extra site kits prior to each site visit to ensure sufficient back-up supplies are available.

3.1.4 Chill Enterococci Filter Extraction Tubes

In preparation for later filtering, remove Enterococci filter extraction tubes with beads from the filter kit provided in the site kit and place on dry ice.

3.2 Post Sampling Activities

Upon return to the launching location after sampling, the team must perform a post-measurement calibration check of the multi-probe meter, review all completed data forms and labels for accuracy, completeness, and legibility and make a final inspection of samples. If obtainable samples are missing, the site should be rescheduled for complete sampling. Other post sampling activities include: inspection and cleaning of sampling equipment, supply inventory, sample and data form shipment, and communications.

3.2.1 Post-measurement Calibration Check of Multi-probe Meter

After all *in situ* measurements have been completed for the sampling day, the team must perform a post-measurement calibration check of the instrument to detect any drift from the pH and conductivity calibration values. To do this, measure the pH and conductivity of the

respective calibration standards that were used earlier in the day to calibrate the instrument. Record these values on the Field Measurement Form. If significant drift is detected, it may indicate that the meter is in need of service.

3.2.2 Review Data Forms and Labels

The field crew leader is ultimately responsible for reviewing all data forms and labels for accuracy, completeness, and legibility. Ensure that written comments use no "shorthand" or abbreviations. The data forms must be thoroughly reviewed. If information is missing from the forms or labels, the Field Team Leader is to provide the missing information. Upon completing the review, the field crew leader must initial the field forms to indicate that they are ready to be sent to the Information Management Center. Each sample label must also be checked for accuracy, completeness, and legibility. The field crew leader must cross-check the sample numbers on the labels with those recorded on the data forms and tracking forms.

3.2.3 Inspect and Prepare Samples

All samples need to be inspected and appropriately preserved and packaged for transport. Check that all samples are labeled, and all labels are completely filled in. Check that each label is covered with clear plastic tape. Check the integrity of each sample container, and be sure there are no leaks. Make sure that all sample containers are properly sealed. Make sure that all sample containers are properly preserved for storage or immediate shipment.

3.2.4 Equipment Cleanup and Check

All equipment and gear must be cleaned and disinfected when traveling over land between distinctively different sites to reduce the risk of transferring nuisance species and pathogens. Species of primary concern in the U.S. include zebra mussels (*Dreissena polymorpha*), mitten crabs (*Eriocheir sinensis*), and Eurasian ruffe (*Gymnocephalus ceinuus*). Field crews must be aware of regional species of concern, and take appropriate precautions to avoid transfer of these species. There are several online resources regarding invasive species, including general information about aquatic nuisance species available from the Aquatic Nuisance Species Task Force (http://www.anstaskforce.gov). General information about freshwater invasive species is available from the U.S. Geological Survey Nonindigenous Aquatic Species website (http://nas.er.usgs.gov), the *Protect Your Waters* website that is co-sponsored by the U.S. Fish and Wildlife Service (http://www.protectyourwaters.net/hitchhikers), and the Sea Grant Program (http://www.sgnis.org). In the Great Lakes, Viral Hemorrhagic Septicemia (VHS) is a deadly fish virus and an invasive species that is threatening Great Lakes fish. VHS was diagnosed for the first time ever in the Great Lakes as the cause of large fish kills in lakes Huron, St. Clair, Erie, Ontario, and the St. Lawrence River in 2005 and 2006.

Handle and dispose of disinfectant solutions properly, and take care to avoid damage to lawns or other property. Table 3-3 describes equipment care. Inspect all equipment, including nets, boat, and trailer, and clean off any plant and animal material. Prior to leaving a site, drain all bilge water and live wells in the boat. Inspect, clean, and handpick plant and animal remains from vehicle, boat, motor, and trailer. Before moving to the next site, if a commercial car wash facility is available, wash vehicle, boat, and trailer and thoroughly clean (hot water pressurized rinse--no soap). Rinse equipment and boat with 1% - 10% bleach solution to prevent the spread of exotics. For specific techniques to disinfect boats and gear in the Great Lakes, please see Appendix C.

Table 3-3. Postsampling Equipment Care

1. Clean for biological contaminants.

- Prior to departing site, drain all bilge water from the boat.
- Inspect motor, boat, trailer, sampling gear, foul weather gear, boots, etc. for evidence of mud, snails, plant fragments, algae, animal remains, or debris, and remove using brushes or other tools.
- At the base location, inspect and rinse seines, dip nets, sieves, foul weather gear, and boots with water and dry. Use one of the procedures below to disinfect gear if necessary.

Additional precautions to prevent transfer of Whirling Disease spores, New Zealand mudsnails, and amphibian chytrid fungus is important for Great Lakes sites. Before visiting the site, research the site and determine if it is in an area where whirling disease, New Zealand mud snails, or chytrid fungus are known to exist. Contact the local State fishery biologist to confirm the existence or absence of these organisms.

- If the site is listed as "positive" for any of the organisms, or no information is available, *avoid using felt-soled wading boots*, and, after sampling, disinfect **all** fish and benthos sampling gear and other equipment that came into contact with water or sediments (i.e., waders, boots, etc.) by one of the following procedures:

  Option A:

  1. Soak gear in a 10% household bleach solution for at least 10 minutes, or wipe or spray on a 50% household bleach solution and let stand for 5 minutes
  2. Rinse with deionized water (do not use sea or lake water), and remove remaining debris
  3. Place gear in a freezer overnight, soak in a 50% solution of Formula 409® antibacterial cleaner for at least 10 minutes or soak gear in 120°F (49°C) water for at least 1 minute.
  4. Dry gear in direct sunlight (at least 84 °F) for at least 4 hours.

  Option B:

  1. Soak gear in a solution of Sparquat® (4-6 oz. per gallon of water) for at least 10 minutes (Sparquat is especially effective at inactivating whirling disease spores).
  2. Place gear in a freezer overnight or soak in 120°F (49°C) water for at least 1 min.
  3. Dry gear in direct sunlight (at least 84 °F) for at least 4 hours.

2. Clean and dry other equipment prior to storage.

- Rinse coolers with water to clean off any dirt or debris on the outside and inside.
- Make sure water quality meter probes are rinsed with deionized water and stored moist.
- Rinse all beakers used to collect water chemistry samples three times with deionized water. Place sampling equipment in a clean location for use at the next site.
- Check nets for holes and repair or locate replacements.

3. Inventory equipment and supply needs and relay orders to the Field Logistics Coordinator.

4. Remove GPS and multi-probe meter, and set up for predeparture checks and calibration. Examine the oxygen membranes for cracks, wrinkles, or bubbles. Replace if necessary, allowing sufficient time for equilibration.
5. Recharge/replace batteries as necessary.
6. Replenish fuel and oil;
7. If a commercial car wash facility is available, thoroughly clean vehicle and boat (hot water pressurized rinse—no soap).

3.2.5 Shipment of Samples and Forms

3.2.5.1 Samples

The field team must ship or deliver time-sensitive samples (i.e., water chemistry, chlorophyll-*a,* sediment-grain size, sediment-TOC, and dissolved nutrients) to the appropriate analytical laboratory (WRS Corvallis) as soon as possible after collection. Other samples (see Appendix D) may be shipped or delivered in batches provided they can be adequately preserved. Batched samples should be shipped every two weeks. Field teams are to fill out one sample tracking form for each sample shipment. On each sample tracking form, the following information must be recorded:

- Airbill or package tracking number;
- Date sample(s) were sent;
- Site ID where each sample was collected;
- Sample type code:

CHEM – Water Chemistry
CHLA – Chlorophyll-*a*
NUTS – Dissolved Nutrients
ENTE – Enterococci
FTIS – Fish Tissue Composite
PACK – Field Form Packet
HTIS – Human Health Fish Tissue (Great Lakes subset only)
BENT – Benthos Grab
SEDX – Sediment Toxicity
SEDG – Sediment Grain Size
SEDO – Sediment Organics/Metals
SEDC – Sediment TOC
PHYT – Phytoplankton (Great Lakes Only)
UVID – Underwater Video (Great Lakes Only)

- Date when the sample(s) was collected (1st day if sampling took >1 day);
- Site visit number (e.g., 1 for first visit, 2 for re-visit);
- Sampler name and contact information;
- Sample ID number encoded on label;
- Number of containers for each sample;
- Destination lab; and
- Any additional comments.

It is important that the correct tracking form (see Section 3.2.6.2) be completed, placed into a waterproof plastic bag, and secured inside the shipping container (e.g., taped to the inside top of the coolers). **Detailed sample shipping instructions are presented in Appendix D.**

3.2.5.2 Field Forms

After checking the Field Forms for completeness and accuracy, the Field Crew Leader will make copies of all Field Forms and retain the copies. **The original forms will be mailed to Marlys Cappaert in the FedEx envelope provided in the site kit.** A pre-addressed airbill will be provided. The original forms must be sent because they are printed specifically to be used in a scanner for automated data entry. Field forms may be retained and mailed in batches throughout the field season (every 2 weeks) when it is convenient to make the copies. **All field forms must be turned in within 2 weeks of completing sampling.** A tracking form will be submitted with each shipment of data forms. The data forms will be tracked in the same manner as all other samples.

3.2.6 Status Reports and Communications

3.2.6.1 Sample Status Report

After each site visit, the field crew leader must submit a sample status report to the Information Management Coordinator to report the site visit and indicate which samples were collected and their respective sample ID numbers. This status report must be submitted before the time sensitive samples are due to arrive at the lab. This ensures that the laboratory is prepared to receive the samples and will help track down the samples should they become lost in transit. The field crew must also ship all time sensitive samples such that they will arrive at the WRS lab within 48 hours of collection. This requires the samples be sent by Priority Overnight shipping the same day as collection, or at the latest, the following day. **A tracking form must accompany every sample shipment**.

For convenience, the WRS sample tracking form and the site status report are incorporated into a single form called the Tracking and Sample Status form (Figure 3-2). Teams have the option of filling this form out digitally using Microsoft Word or manually filling out the scannable version of the form that is provided in the set of field forms. See below for more information on the form submission options available to crews.

If the crew visits a site with the intention of sampling, but determines the site to be unsampleable (either temporarily or permanently), the site status portion of the form needs to be completed and submitted, but the WRS tracking portion of the form can remain blank.

3.2.6.2 Tracking Forms

Regardless of the type of sample being shipped, a tracking form must be completed for every shipment and a copy must be placed inside the container with the samples (typically sealed in a plastic bag and taped to the inside of the cooler lid). Again, teams have the option of filling this form out digitally using Microsoft Word or manually filling out the scannable version of the form that is provided in the set of field forms. A copy of the tracking form must also be submitted to the Information Management Coordinator before the samples are due to arrive at the lab. See below for more information on the form submission options available to crews.

Samples Shipped Immediately
For time sensitive samples, fill out the Tracking and Sample Status form, the lower portion has the sample ID numbers for each sample being sent.

Batched Samples
Samples that are less time sensitive can be held for up to 2 weeks and sent to the appropriate lab in batches. Fill out a Batched Samples Tracking Form (Figure 3-3) with the sample types and sample ID numbers. **Use a separate form for each lab.**

This form must be filled out and submitted:

- when samples are brought into your lab or holding facility, and
- then again when the samples are shipped.

Fish Tissue and Human Health Fish Tissue Samples
Separate tracking forms will be completed and submitted for each fish tissue sample. Separate tracking forms are used for the ecological fish tissue collections (Figure 3-4) and human health fish tissue collections (Figure 3-5). Human health fish tissue samples will be collected at a subset of Great Lakes sites only.

3.2.6.3 Methods of Communication (in order of preference)

1) **E-mail submission:** Electronic versions of the Tracking and Sample Status form and the Batched Samples Tracking form will be emailed to the field crew leaders after their training session. Complete the Tracking and Sample Status report form for each site, even sites that are visited but not sampled, and email the form to sampletracking@epa.gov. This email will go to both the Information Management Coordinator and the Field Logistics Coordinator.

   You must follow a standardized naming convention when naming the electronic Tracking and Sample Status report files. The naming convention for this form is:
   "labid_siteid_datecollected.doc:"

   ex. WRS_NCCA10OR123_06_28_2010.doc

   For batch samples, the naming convention is:
   "BR_labid_siteid_datecollected.doc:"

   ex. BR_GLEC_NCCA10OR123_06_28_2010.doc (*in this case, the site id and date collected will refer to the first sample on the page*)

   Be sure to print a copy of the tracking form to be placed inside the container with the samples prior to sealing the container for shipping.

2) **Fax Submission:** If you are not able to email the electronic forms, the forms can be faxed on a **non**-voice over internet phone (VOIP) fax machine. This could be a printed version of your electronic form or a hand written copy of the scannable version of the form that is provided in the set of field forms.
   The fax number for tracking form submission is 541-754-4637

3) **Voice Submission**: If neither emailing nor faxing the forms is feasible, the team leader may call into the number provided on the bottom of the Tracking and Sample Status form (read the ENTIRE form to the voice mail machine. It is extremely important that all information from the form is transferred; do not leave any information out of your message). **The voice number for tracking form submission is 541-754-4663**

Information Management Coordinator

Marlys Cappaert	541-754-4467	Cappaert.Marlys@epamail.epa.gov

Sample Tracking email submission:	sampletracking@epa.gov
Sample Tracking fax submission:	541-754-4637
Sample Tracking voice submission:	541-754-4663

Primary Field Logistics Coordinator

Jennifer Linder	410-356-8993	Jennifer.Linder@tetratech.com

Alternate Field Logistics Coordinator

Chris Turner	715-829-3737	cturner@glec.com

Other Information

It is very important to complete the sample status report **immediately after every sampling event**. This will enable the Field Logistics Coordinator to track sampling progress. More importantly, it will enable the Information Management Center to track which samples were collected at each site, and to immediately track the shipment of the time-sensitive samples that will be shipped after each sampling event.

The field crews should call or email the Field Logistics Coordinator to report any problems encountered. The Field Logistics Coordinator monitors all aspects of field sampling activities. The Field Logistics Coordinator and Information Management Coordinator will contact the EPA Headquarters Coordinator regularly to provide regional updates throughout the sampling period. The EPA Headquarters Coordinator will maintain a database of all sampling activities and reconnaissance information.

The EPA Regional Coordinator serves as the central point of contact for information exchange among field teams, the management and QA staffs, the information management team, and the public. A list of EPA Regional Coordinators and their contact information can be found at the beginning of this manual on page ix.

NCCA 2010 TRACKING AND SAMPLE STATUS - WRS

SITE ID: NCCA10- Visit #: ○ 1 ○ 2 Date Collected: ___/___/2010

SENT BY: _____ SENDER PHONE: _____

State of Site Location: _____ TEAM: _____ DATE SENT: ___/___/2010

SHIPPED BY: ○ FedEx ○ UPS ○ Hand Delivery ○ Other: _____ AIRBILL/TRACKING NUMBER: _____

SITE STATUS			SAMPLE STATUS
			○ No Samples Collected
SAMPLEABLE	NOT SAMPLEABLE	NO ACCESS	Mark the samples that were collected during this site visit:
○ Marine ○ Great Lakes Temporarily Not Sampleable ○ Other (Explain in Status Comments)	○ Shallow ○ Unsafe ○ Map Error ○ Other (Explain in Status Comments)	○ Access Denied ○ Inaccessible ○ Temp Inaccessible	○ Water Chem (CHEM) ○ Sediment TOC (SEDC) ○ Chlorophyll-a (CHLA) ○ Sediment Grain (SEDG) ○ Nutrients (NUTS) ○ Benthos Grab (BENT) ○ Sediment Organics (SEDO) ○ Enterococci (ENTE) ○ Sediment Toxicity (SEDX) ○ Fish Tissue (FTIS) (Great Lakes Only) ○ Human Health Fish Tissue (HTIS) ○ Video Card (UVID) ○ Phytoplankton (PHYT)

Status Comments

Sample ID	Sample Type	Comments
	CHEM	
	CHLA	
	NUTS	
	SEDC	
	SEDG	

Sample Types	Condition Codes	Chain of Custody	Contact Information
CHEM - Water chemistry CHLA - Chlorophyll-a NUTS - Nutrients SEDC - Sediment TOC SEDG - Sediment Grain Size	Filled in by recipient C = Cracked jar F = Frozen L = Leaking ML = Missing label NP = Not preserved W = Warm OK = Sample OK T = Thawed	Filled in by recipient Date Received: ___/___/___ Received by: _____	Tracking Assistance: Marlys Cappaert 541-754-4467 Michelle Gover 541-754-4793 Lab: Attn: Phil Monaco, Dynamac c/o U.S. EPA 1350 Goodnight Ave Corvallis, OR 97333 PH: 541-754-4720 monaco.phil@epamail.epa.gov

FAX THIS FORM TO 541-754-4637
OR READ TRACKING INFO TO VOICE MESSAGE CENTER:
541-754-4663

04/22/2010 NCCA Tracking - WRS 8299111157

Figure 3-2. Example Tracking and Sample Status Form

NCCA 2010 TRACKING - BATCHED SAMPLES

SENT BY: _____ SENDER PHONE: _____ STATE OF SITE LOCATION: _____ TEAM: _____

Shipped By: O FedEx O UPS O Hand Delivery DATE SHIPPED: ___ / ___ / 2010

AIRBILL/TRACKING NUMBER: _____

O Held at address: _____

Site ID	Date Sample Collected MM/DD/YYYY	Visit	Sample ID	Sample Type	# of Containers	Comments	Cond. Code
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					
NCCA10-		O 1 O 2					

Lab

- O BENTHIC LAB
- O GLEC
- O MED
- O NERL - N. CHELMSFORD, MA
- O TETRA TECH
- O WED - CORVALLIS, OR
- O OTHER _____

Chain of Custody

Filled in by recipient

Date Received: ___/___/___

Received by: _____

Sample Types

BENT - Benthos Grab
ENTE - Enterococci
SEDX - Sediment Toxicity
SEDO - Sediment Organics/Metals
PHYT - Phytoplankton (Great Lakes Only)
PACK - Field Form Packet

Condition Codes

Filled in by recipient

C = Cracked jar
F = Frozen
L = Leaking
ML = Missing label
NP = Not preserved
W = Warm
OK = Sample OK
T = Thawed

Tracking Assistance:

Marlys Cappaert 541-754-4467
Michelle Gover 541-754-4793

FAX THIS FORM TO 541-754-4637 OR READ TRACKING INFO TO VOICE MESSAGE CENTER: 541-754-4663

04/22/2010 NCCA Tracking - Batch

1059022391

Figure 3-3. Example Batched Samples Tracking Form

NCCA 2010 ECO FISH TISSUE TRACKING

SITE ID: NCCA10- VISIT: ○ 1 ○ 2 DATE COLLECTED: ___ / ___ / 2010

SENT BY: SENDER PHONE:

STATE OF SITE LOCATION: TEAM: SHIPPED BY: ○ FedEx ○ Other:

AIRBILL/TRACKING NUMBER: DATE SHIPPED: ___ / ___ / 2010

SAMPLE ID

	Genus Species	Total Length (mm)	Comments	Condition Code
.01				
.02				
.03				
.04				
.05				
.06				
.07				
.08				
.09				
.10				
.11				
.12				
.13				
.14				
.15				
.16				
.17				
.18				
.19				
.20				

Filled in by recipient

Condition Codes

L = Leaking
ML = Missing label
NP = Not preserved
W = Warm
OK = Sample OK
T = Thawed

Chain of Custody

Date Received:
____/____/ _____

Received by:
_____

Contact Information

Tracking Assistance:
Marlys Cappaert
541-754-4467

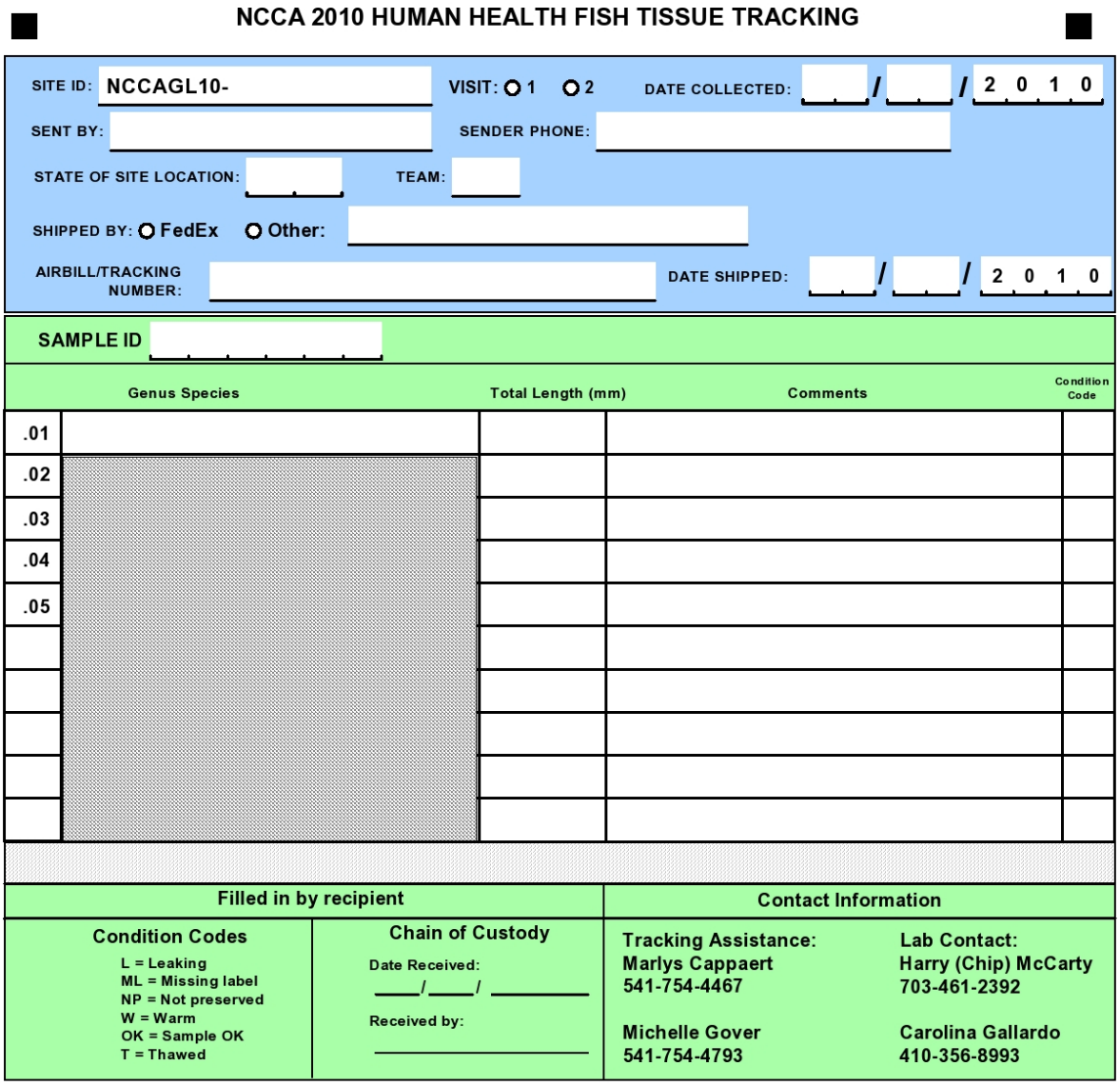
Michelle Gover
541-754-4793

Lab Contact:
???
PH: ???

04/22/2010 NCCA Tracking - Fish Tissue

3080542800

Figure 3-4. Example Ecological Fish Tissue Tracking Form



NCCA 2010 HUMAN HEALTH FISH TISSUE TRACKING

SITE ID: NCCAGL10- VISIT: ○ 1 ○ 2 DATE COLLECTED: ___/___/2010

SENT BY: SENDER PHONE:

STATE OF SITE LOCATION: TEAM:

SHIPPED BY: ○ FedEx ○ Other:

AIRBILL/TRACKING NUMBER: DATE SHIPPED: ___/___/2010

SAMPLE ID

	Genus Species	Total Length (mm)	Comments	Condition Code
.01				
.02				
.03				
.04				
.05				

Filled in by recipient

Condition Codes

L = Leaking
ML = Missing label
NP = Not preserved
W = Warm
OK = Sample OK
T = Thawed

Chain of Custody

Date Received: ____/____/ _____

Received by: _____

Contact Information

Tracking Assistance:
Marlys Cappaert
541-754-4467

Michelle Gover
541-754-4793

Lab Contact:
Harry (Chip) McCarty
703-461-2392

Carolina Gallardo
410-356-8993

04/22/2010 NCCA Tracking - Human Health Fish Tissue

5582497224

Figure 3-5. Example Human Health Fish Tissue Tracking Form

4.0 INITIAL SITE PROCEDURES

When you arrive at a site, you must first confirm you are at the correct site, and then determine if the site meets the criteria for sampling and data collection activities (see Site Evaluation Guidelines). Inspect the site for appropriate access, safety, and general conditions to determine if the site can be sampled within the swing of the boat while anchored.

Please remember that crew members responsible for collecting water chemistry, sediment and fish tissue samples must not apply sunscreen or other chemical contaminants until after the sample is collected.

4.1 Site Verification Activities

4.1.1 Locating the X-Site

Base site sampling points were chosen using a Generalized Random Tessellation Stratified (GRTS) survey design, which is a stratified design with equal probability of selection. Each point is referred to as the "X-site." The X-site is the point that determines the location at which samples are taken. The latitude/longitude of the X-site is listed on the site spreadsheet that was distributed by the EPA Regional Coordinators. Table 4-1 provides a list of equipment and supplies necessary for locating and verifying the sampling site.

4.1.2 Target population

The target population for the estuarine resources consists of all coastal waters of the conterminous United States from the head-of-salt to confluence with the ocean, including inland waterways, tidal rivers and creeks, lagoons, fjords, bays, and major embayments such as Florida Bay and Cape Cod Bay (see Figure 4-1 and 4-2). For the purposes of this study, the head of salt is generally defined as < 0.05 psu (ppt) and represents the landward/upstream boundary. The seaward boundary extends out to where an imaginary straight-line intersecting two land features would fully enclose a body of coastal water. All waters within the enclosed area are defined as estuarine, regardless of depth or salinity.

The target population for the Great Lakes encompasses all near-shore waters. The near shore zone is defined as the region from the shoreline to 30m in depth constrained to a maximum of 5 km from shore. The Great Lakes include Lake Superior, Lake Michigan, Lake Huron, Lake Erie, and Lake Ontario. The National Aquatic Survey will be restricted to the United States portion.

Table 4-1. Equipment and Supplies List for Site Verification.

For locating and verifying site	▪ Sampling permit and landowner access (if required) ▪ Field Operations Manual and/or laminated quick reference guide ▪ Site packet, including access information, site spreadsheet with map coordinates, navigational charts with "X-site" marked ▪ NCCA Fact Sheets for public outreach ▪ GPS unit (preferably one capable of recording waypoints) with manual, reference card, extra battery pack
For recording measurements	▪ Clipboard ▪ #2 pencils ▪ Site Verification Form

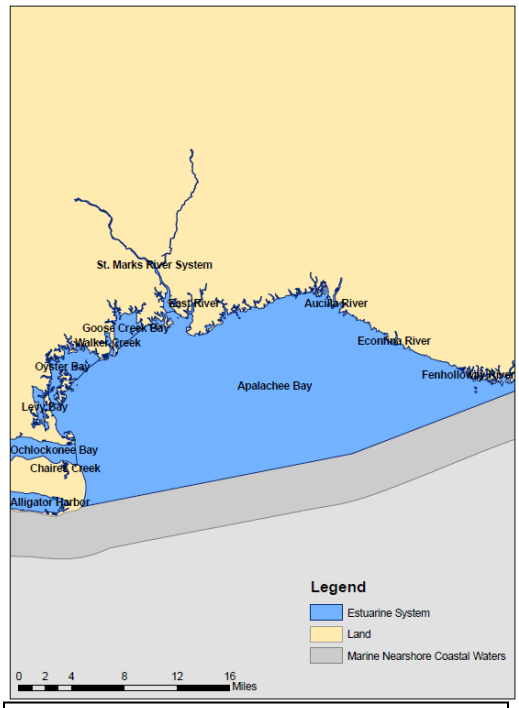


Figure 4-1: Example of an Estuarine System Comprised of an Embayment plus a Complex of Bays and Tidal Rivers and Creeks.

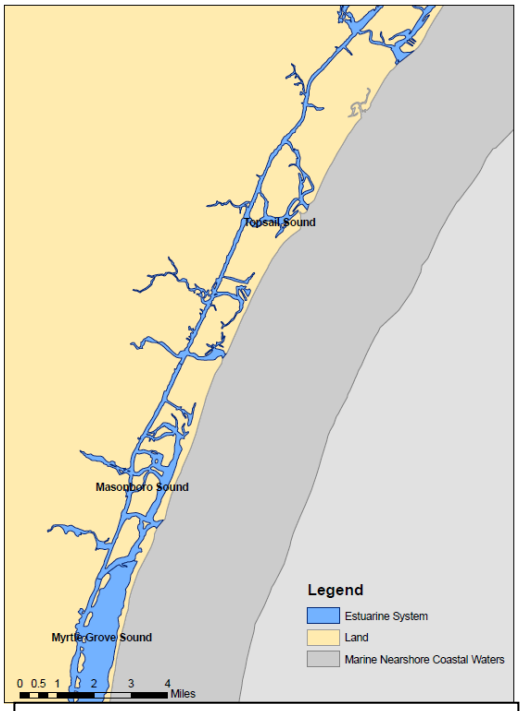


Figure 4-2: Example of an Inter-coastal Estuarine System.

4.1.3 Site Verification

Identify the X-site using GPS and navigate to the location within 0.02 nautical miles (nm) (±37 meters) of the given coordinates. Record the coordinates from the EPA-provided site list on the field verification form as the "target" coordinates. Record the actual coordinates of the vessel after anchorage on the field verification form as the "actual" coordinates. Samples should be taken as close to these coordinates (actual X-site) as possible. Teams are expected to collect all samples within a circle of 0.02 nm radius around the X-site (see Section 4.1.6 regarding secondary collection areas for sediment). This distance should account for typical "anchor swing" of the sampling vessel. It is recommended that teams create a GPS waypoint shortly after anchoring the boat and periodically consult the GPS during sampling to ensure that the sampling vessel has not drifted away from the X-site due to anchor drag.

The field team must verify that the site is correctly located. Sampling site verification is based on map coordinates and locational data from the GPS. Record locational coordinates for the site on the Site Verification Form. Latitude and longitude are required for all sampled sites. Include the number of satellites fixed (≤ 3 or ≥ 4), and the GPS datum used (e.g., WGS 84) for QA purposes. Compare the map coordinates given on the coastal survey spreadsheet for the targeted sampling point with the GPS coordinates displayed for the sampling site, and check to see if the two sets of coordinates are within 0.004167 decimal degrees of latitude and longitude. This distance is approximately equal to the precision of the GPS receiver without differential correction of the position fix. This is the desired level of precision.

Size and shape of the waterbody in which the X-site resides will vary depending on coastal morphology. While site depth or salinity should not generally be a factor for abandoning a planned sampling site, there are some points that a field crew may consider in order to determine if an X-site meets the operational definition of an estuary in marine environments, or lacustrine and near shore coastal waters in the Great Lakes:

- Access to open water;
- Navigable using a shallow-draw boat. Typically this means that the depth of the X-Site is generally ≥ 1 meter. Actual depth determinate, however, may be based on the vessel and sampling equipment used, and wave action at the site.

If the specific site does not fit this definition and every attempt to relocate a site within the margin provided (see Section 4.1.4) has been made, complete the appropriate "Non-Sampleable-Permanent" category on the Site Verification Form and provide an explanation for not sampling the site in the comments section of the form. Add any additional explanation as required. (For complete details on the site evaluation process, refer to the Site Evaluation Guidelines).

If the site is non-sampleable or inaccessible, no further sampling activities are conducted. Replace the site with the next oversample site on the estuary/state list. Notify the EPA Regional Coordinator and Field Logistics Coordinator (Section 3.2.6) that the site was replaced and submit the verification form to the Information Management Coordinator. Refer to the See Site Evaluation Guidelines for further information regarding the replacement of sites.

If the site is sampleable, record the sampling status and pertinent site verification information on the Site Verification Form (Figure 4-3) including the time of arrival at the X-site and water depth. Make sure an accurate depth reading is taken at the site to ensure the depth is adequate to conduct sampling. Refer to Table 4-2 for detailed information regarding the site verification process.

Record the names of the field team on the back of the Site Verification Form (Figure 4-4). Also create a general site sketch of the area. Include the relative locations of the shoreline, launch point, X-site, fish collection location, and sediment location if different than the X-site (see Section 4.1.6). Include any other specific attributes of the site that may be important during the data processing portion of the assessment. A printed or copied section of a map with the pertinent information may be submitted in place of the site sketch.

4.1.4 Site Relocation

For relocation situations, every attempt should be made to relocate a site within a 0.02 nm (~37 m) radius of the intended location. A guideline for the relocation effort follows:

- The field crew leader should choose a specific compass heading (e.g., north, south, east, west) and slowly motor the vessel in that direction for approximately 15-20 meters. Assess the relocated site using the Site Verification guidelines given above. Should the relocated site fail to meet the operational definition "sampleable", then this process may be continued using the same heading out to the 37 meter mark or using a new heading until an acceptable sampling location is found. If after a sufficient amount of effort is expended and no suitable site is found, then the determination may be made that the site is unsampleable.

4.1.5 Sample Collection

While the field crew should make every attempt to collect all samples, there will be some circumstances that will prevent this from happening. Following is a guideline for prioritizing sample collection at each site and a "check-list" for determining sample completion when site conditions limit full completion of the standard sampling protocol:

- In-situ water measurements and water samples are expected to be collected at all sites and in their entirety;
- Benthic samples should always be collected. Any sediment size is acceptable so long as the definition of a "successful grab" is met (see Section 6.1.3);
- Sediment composite material of sand-sized sediment grain or smaller, should be collected;
    - Since there may be cases where only a limited amount of sediment can be acquired for the sediment chemistry, characterization, and toxicity composite, the listing given below provides the expected sample in order of preference:
        1) Organics/Metals
        2) Total Organic Carbon (TOC)
        3) Silt/Clay (Grain size)
        4) Toxicity
    - Flag and note the reason for limited sediment sample collections;
- Fish collection for ecological contaminant analysis. For the ecological assessment, fish collection may be conducted anywhere within a 500 m radius of the X-Sites. Successful deployment of fish collection gear or the absence of fish in the catch should not necessarily be used as a determining factor for rendering a site "unsampleable". Human health fish sampling (subset of Great Lakes sites) requirement is covered separately.

4.1.6 Secondary Sediment Collection Areas

All water, benthos and sediment samples are expected to be collected at the same location, as close to the X-Site as possible. However, there may be circumstances that arise that require the field crew to relocate in order to successfully complete sampling at a site; specifically for sediment samples. If benthos and/or sediment are collected from a secondary location, in situ measurements and water collections do not need to be resampled. For these situations, a guideline for locating a secondary sediment sample collection area follows:

- If an acceptable sediment grab can not be obtained at the X-site, it is permissible to move around within the 37 meter margin (of the X-site) to obtain the sediment sample, using the site relocation method described previously (this is still considered the primary sediment sample collection area). Mark the circle on the Sample Collection Form indicating that sediment was collected within 37 meters of the X-site. *Acceptable* means:
    1) A sediment grab that meets the criteria for benthic samples; or
    2) Enough sediment can be collected that will allow the crew to collect the sediment surface sub-sample required for the sediment composite that will be used to send to send to the laboratory for abiotic indicator analysis (e.g., organics/metals, TOC, grain size, toxicity).
- In cases where sediment sampling can be successfully conducted in a secondary sediment collection area (e.g., > 37 m radius but within a 100 m radius (~0.05 nm) of the X-site), then the field team may do so without having to re-collect the water samples.

Mark the circle on the Sample Collection Form indicating that sediment was collected between 37 and 100 meters from the X-site. Indicate in the comments section approximately how far from the X-site and in what direction the sediment was collected. Indicate the relative location of the sediment collection on the map on the back of the Site Verification Form. The data will be flagged for subsequent review.

- **In Great Lakes sites only**, teams may move out to a maximum distance of 500 meters from the X-site in repeated attempts to locate suitable sediment sampling locations (after attempting to collect sediment from within the primary and secondary locations). Mark the circle on the Sample Collection Form indicating that sediment was collected between 100 and 500 meters from the X-site. Indicate in the comments section approximately how far and in what direction the sediment was collected. Indicate the relative location of the sediment collection on the map on the back of the Site Verification Form. The data will be flagged for subsequent review.
- If a suitable location to collect sediment samples has not been found after a minimum of 3 collection attempts inside each of the acceptable relocation areas, then sampling is considered "complete" for the site. All appropriate field form flags and explanations should be filled in. NOTE: More sediment grab attempts may be completed at the discretion of the field team leader.

4.1.7 Habitat Assessment

Indicate on the site verification form the basic habitat type and bottom composition present at the X-site. Note the presence and type of any marine debris, submerged aquatic vegetation (SAV), and/or macroalgae present in the area.

4.1.8 Site Photograph

Taking site photographs is an optional activity that is encouraged, but should be considered if the site has unusual natural or man-made features associated with it. If you do take photographs with a digital camera at a site, date-stamp the photograph and include the site ID. Alternatively, start the sequence with one photograph of an 8.5 × 11 inch piece of paper with the site ID, waterbody name, and date printed in large, thick letters. After the photo of the site ID information, take any additional photos you find interesting after this first picture. Keep a log of your photographs and briefly describe each one. EPA encourages crews to submit digital pictures to the regional and headquarters contacts for use in NCCA assessment materials.

Reviewed by (initial): JPS

NCCA 2010 SITE VERIFICATION (Front)

SITE NAME: GULF OF MEXICO DATE: 06/01/2010 VISIT: ● 1 ○ 2

SITE ID: NCCA10 LA0000 STATE OF SITE LA STATION DEPTH(m): 6.0 TEAM: LA1

DID YOU SAMPLE THIS SITE?

● YES If YES, check one below

SAMPLEABLE (Choose method used)
● Marine
○ Great Lakes

ARRIVAL TIME: 07:45
DEPART TIME: 13:30

○ NO If NO, check one below

NON-SAMPLEABLE-PERMANENT-Replace Site
○ Map Error
○ Site too shallow for navigation/sampling
○ Unsafe

NON-SAMPLEABLE-TEMPORARY-Reschedule
○ No Access
○ Temporarily Inaccessible-Fire, etc.
○ Other (Explain in comments)

VERIFICATION INFORMATION

Site verified by (fill in all that apply): ● GPS ○ Local Contact ○ Signs ○ Roads ○ Topo. Map
○ Other (Describe Here): ○ Not Verified (Explain in Comments)

LOCATION

Coordinates	Latitude North	Longitude West	# of Satellites	X-SITE WITHIN 37M?
TARGET Decimal Degrees	29.722980	091.928652	○ ≤3	● YES ○ NO
ACTUAL Decimal Degrees	29.722970	091.928660	● ≥4	GPS Datum Used (e.g. NAD83,WGS84): WGS84

HABITAT TYPE: ○ Tidal River ○ Open Water ○ Marsh/Wetland ● Embayment ○ Inter-Tidal ○ Rivermouth
○ Other, explain:

BOTTOM TYPE: ○ Coral Reef ○ Oyster Bed ○ Grass Bed ○ Sand ○ Rocky/Shell ○ Hardpan ● Mud
○ Other, explain:

Debris Present?: ● YES ○ NO | If Yes, TYPE: ○ Glass ○ Plastic ○ Wood ○ Cans ● Other, explain: RAFT of TRASH AND FLOATING PLANTS

SAV Present?: ○ Yes ● No ABUNDANCE: (Sparse, dense, etc)

Macroalgae Present?: ● Yes ○ No ABUNDANCE: SPARSE (Sparse, dense, etc)

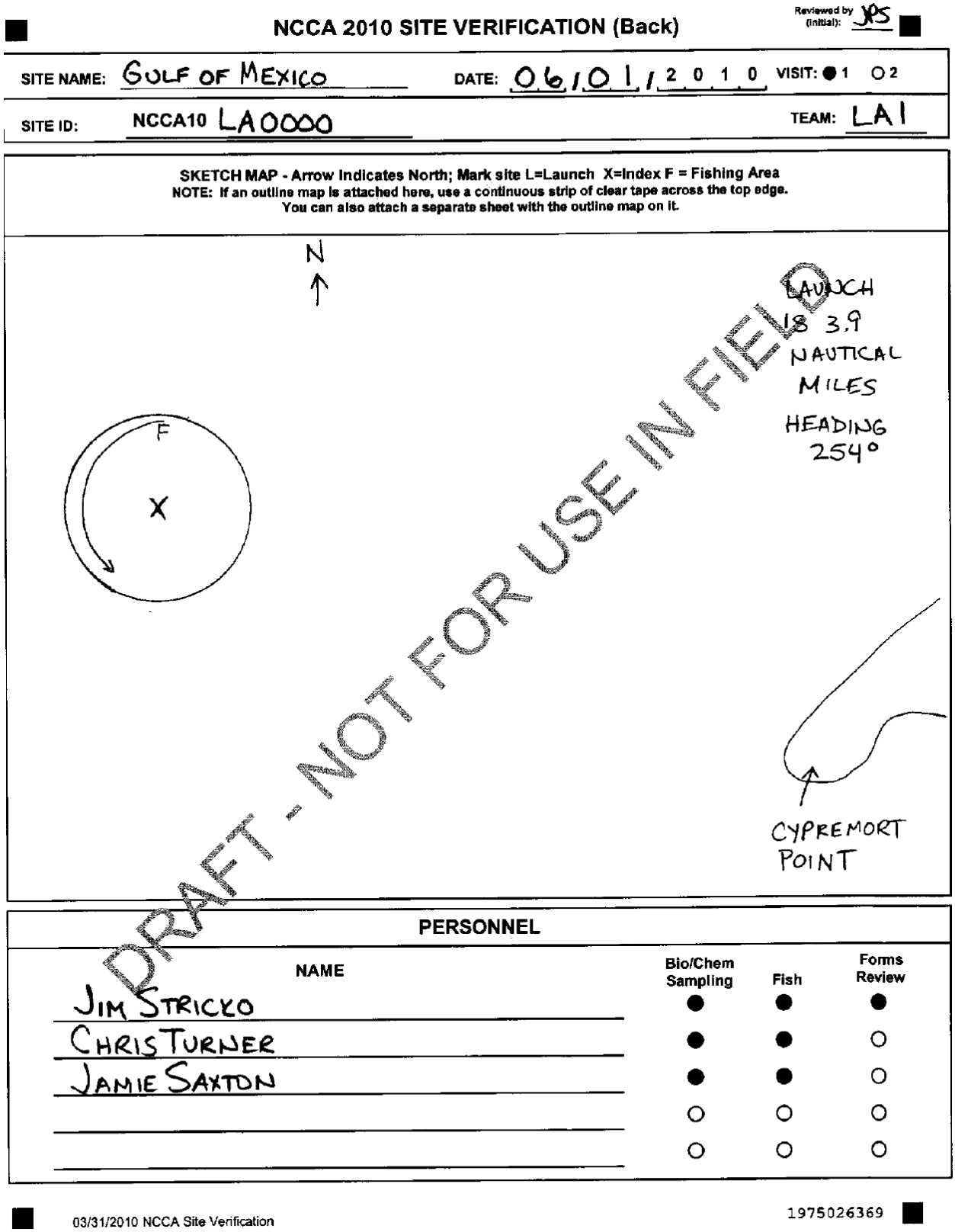
GENERAL COMMENTS: SITE IS LOCATED 2.6 NAUTICAL MILES OFF OF CYPREMORT PORT, LOUISIANA IN VERMILION BAY

DIRECTIONS TO SITE: FROM LA STATE ROUTE 319 - TURN RIGHT ON BEACH LANE. FOLLOW TO CYPREMORT POINT STATE PARK. FOLLOW SIGNS TO BOAT LAUNCH.

03/31/2010 NCCA Site Verification

8194026369

Figure 4-3. Example Site Verification Form (Front)



NCCA 2010 SITE VERIFICATION (Back)

Reviewed by (initial): JPS

SITE NAME: GULF OF MEXICO DATE: 06/01/2010 VISIT: ● 1 ○ 2

SITE ID: NCCA10 LA0000 TEAM: LA1

SKETCH MAP - Arrow Indicates North; Mark site L=Launch X=Index F = Fishing Area
NOTE: If an outline map is attached here, use a continuous strip of clear tape across the top edge.
You can also attach a separate sheet with the outline map on it.

N

LAUNCH
18 3.9
NAUTICAL
MILES
HEADING
254°

F
X

CYPREMORT
POINT

DRAFT - NOT FOR USE IN FIELD

PERSONNEL

NAME	Bio/Chem Sampling	Fish	Forms Review
JIM STRICKO	●	●	●
CHRIS TURNER	●	●	○
JAMIE SAXTON	●	●	○
	○	○	○
	○	○	○

03/31/2010 NCCA Site Verification

1975026369

Figure 4-4. Example Site Verification Form (Back)

Table 4-2. Site Verification Procedures

1. Create a waypoint in the GPS unit that corresponds to the target X-site coordinates provided by EPA in the site list. This can be done ahead of time in the office.
2. Record the coordinates of the target X-site on the Field Verification Form in decimal degrees (Target field).
3. Navigate to the target X-site while noting the distance (in meters) from the given coordinates.
4. Navigate the sampling vessel as close as possible to the target X-site (you must be no more than 0.02 nm or 37 meters from the target X-site).
5. Anchor the sampling vessel in such a way as to minimize the possibility of the anchor(s) dragging or becoming dislodged.
6. Record the time of arrival at the X-site.
7. Record the coordinates of the actual X-site on the Field Verification Form in decimal degrees (Actual field).
8. Create a new GPS waypoint shortly after anchoring and monitor the GPS throughout the sampling to ensure that the sampling vessel has not drifted away from the X-site due to anchor drag.
9. Indicate any and all methods that were used to verify that you are at the correct location.
10. Measure and record the water depth at the X-site on the form.
11. Determine if the site is sampleable - See Site Evaluation Guidelines.
12. If the site is non-sampleable or inaccessible and cannot be relocated within the designated area, indicate the reason on the form. No further sampling activities are conducted. Replace the site with the first oversample site on the estuary/state list. Notify the EPA Regional Coordinator and Field Logistics Coordinator that the site was replaced and submit the verification form to the Information Management Coordinator.
13. If the site is sampleable, record the sampling method being used (marine or Great Lakes).
14. Record the general habitat type and the dominant bottom type present at the sampling site. In many sites, it may not be possible to record the bottom type until after the sediment collections are performed.
15. Record the presence and type any debris or submerged aquatic vegetation (SAV) present.
16. Make any general comments about the site that may be important during the data review portion of the assessment or any unusual characteristics about the site.
17. Record directions to the launch site from an easily recognizable location (city or major road intersection).
18. On the back side of the Field Verification Form, draw a simple sketch of the area. Include the relative locations of the shoreline, launch point, X-site, sediment and fish collection location. Include any other specific attributes of the site that may be important during the data processing portion of the assessment. A printed or copied section of a map with the pertinent information may be submitted in place of the scene sketch.
19. Record the name of crew member and indicate their primary duties.

5.0 WATER QUALITY MEASUREMENTS

5.1 Water Quality

This section describes the procedures and methods for the field collection and analysis of the water quality indicators (*in situ* measurements, water chemistry, Secchi disk transparency, and light attenuation) from freshwater and marine coastal areas.

5.1.1 In Situ Measurements of Dissolved Oxygen, pH, Temperature, Salinity, Conductivity, Transparency, and Light Attenuation

5.1.1.1 Summary of Method

Measure dissolved oxygen (DO), pH, temperature, and salinity (marine sites) or conductivity (freshwater sites) using a calibrated multi-parameter water quality meter (or sonde). Measure water column transparency using a Secchi disk and light attenuation using a Photosynthetically Active Radiation (PAR) meter. Take all the measurements at the X-site at the following depths: 0.1 m below the surface, 0.5 meters below the surface, every 1 meter from depths of 1.0 to 10.0 meters, and every 5 meters thereafter if the site is greater than 10 m. Take the last set of measurements at 0.5 m from the bottom. Be sure the site depth is accurately measured and recorded before taking the water quality measurements. Take care to avoid the probes contacting bottom sediments, as the instruments are delicate. Refer to table 5-3 for detailed instructions on taking the hydrographic profile.

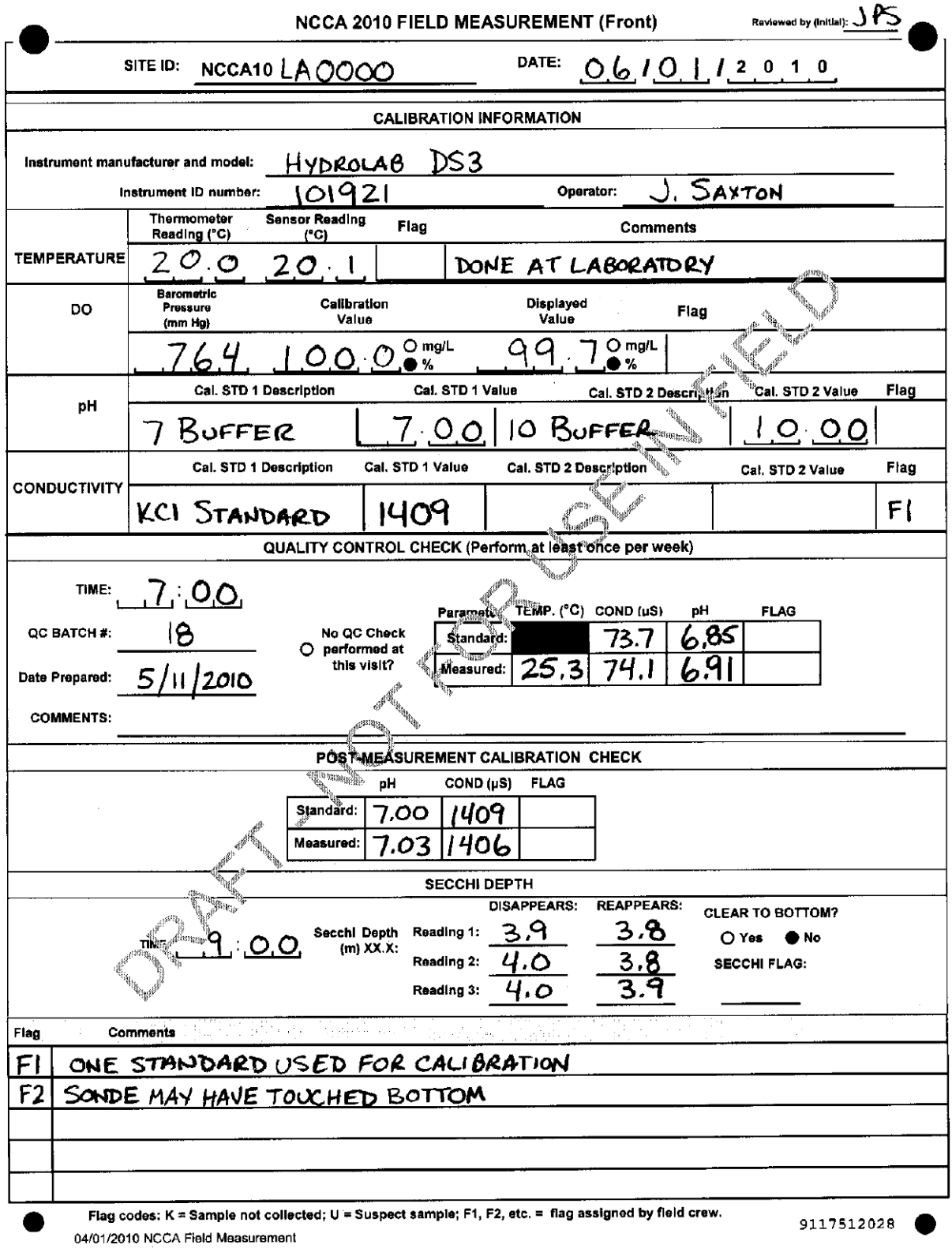
The hydrographic profile will include a full set of measurements on both the downcast (lowering the probe through the water column), and the upcast (as the probe is brought back to the surface). The downcast and upcast measurements will be taken at the same depths.

5.1.1.2 Equipment and Supplies

Table 5-1 provides the equipment and supplies needed to measure transparency, light attenuation, dissolved oxygen, pH, temperature, and salinity/conductivity. Record the measurements on the Field Measurement Form, as seen in Figures 5-1 and 5-2.

Table 5-1. Equipment and Supplies—DO, pH, Temperature, Salinity/Conductivity, Transparency, and Light Attenuation

For taking measurements and calibrating the water quality meter	▪ Multi-parameter water quality meter with pH, DO, temperature, and salinity/conductivity probes. ▪ Extra batteries ▪ De-ionized water (lab certified preferred, but not required) ▪ Calibration cups and standards ▪ QCS calibration standard ▪ Barometer to use for calibration ▪ Thermometer ▪ Secchi disk ▪ PAR meter with independent datalogger
For recording measurements	▪ Field Measurement Form (see Figures 5-1 and 5-2) ▪ Pencils (for data forms)



NCCA 2010 FIELD MEASUREMENT (Front)

Reviewed by (initial): JPS

SITE ID: NCCA10 LA0000 DATE: 06/01/2010

CALIBRATION INFORMATION

Instrument manufacturer and model: HYDROLAB DS3

Instrument ID number: 101921 Operator: J. SAXTON

	Thermometer Reading (°C)	Sensor Reading (°C)	Flag	Comments
TEMPERATURE	20.0	20.1		DONE AT LABORATORY

	Barometric Pressure (mm Hg)	Calibration Value	Displayed Value	Flag
DO	764	100.0 ○ mg/L ● %	99.7 ○ mg/L ● %	

	Cal. STD 1 Description	Cal. STD 1 Value	Cal. STD 2 Description	Cal. STD 2 Value	Flag
pH	7 BUFFER	7.00	10 BUFFER	10.00	

	Cal. STD 1 Description	Cal. STD 1 Value	Cal. STD 2 Description	Cal. STD 2 Value	Flag
CONDUCTIVITY	KCl STANDARD	1409			F1

QUALITY CONTROL CHECK (Perform at least once per week)

TIME: 7:00

QC BATCH #: 18

Date Prepared: 5/11/2010

○ No QC Check performed at this visit?

COMMENTS:

Parameter	TEMP. (°C)	COND (uS)	pH	FLAG
Standard:		73.7	6.85	
Measured:	25.3	74.1	6.91	

POST-MEASUREMENT CALIBRATION CHECK

	pH	COND (µS)	FLAG
Standard:	7.00	1409	
Measured:	7.03	1406	

SECCHI DEPTH

TIME: 9:00

Secchi Depth (m) XX.X:

	DISAPPEARS:	REAPPEARS:
Reading 1:	3.9	3.8
Reading 2:	4.0	3.8
Reading 3:	4.0	3.9

CLEAR TO BOTTOM? ○ Yes ● No

SECCHI FLAG:

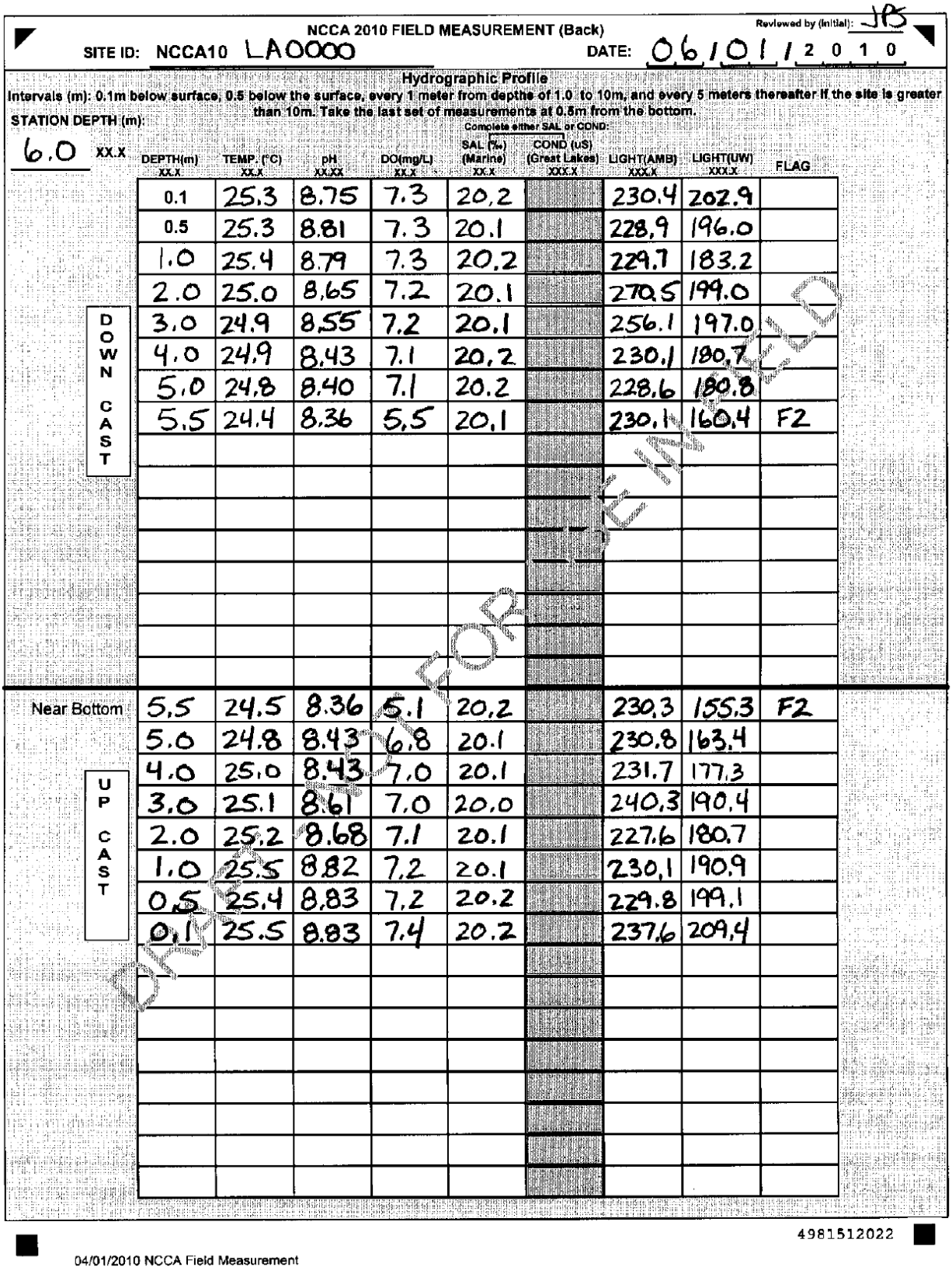
Flag	Comments
F1	ONE STANDARD USED FOR CALIBRATION
F2	SONDE MAY HAVE TOUCHED BOTTOM

Flag codes: K = Sample not collected; U = Suspect sample; F1, F2, etc. = flag assigned by field crew.

04/01/2010 NCCA Field Measurement

9117512028

Figure 5-1. Example Field Measurement Form (Front).



NCCA 2010 FIELD MEASUREMENT (Back)

Reviewed by (initial): JB

SITE ID: NCCA10 LA0000 DATE: 06/01/2010

Hydrographic Profile

Intervals (m): 0.1m below surface, 0.5 below the surface, every 1 meter from depths of 1.0 to 10m, and every 5 meters thereafter if the site is greater than 10m. Take the last set of measurements at 0.5m from the bottom.

STATION DEPTH (m): 6.0 XX.X

Complete either SAL or COND:

	DEPTH(m) XX.X	TEMP. (°C) XX.X	pH XX.XX	DO(mg/L) XX.X	SAL (‰) (Marine) XX.X	COND (uS) (Great Lakes) XXX.X	LIGHT(AMB) XXX.X	LIGHT(UW) XXX.X	FLAG
DOWN CAST	0.1	25.3	8.75	7.3	20.2		230.4	202.9	
	0.5	25.3	8.81	7.3	20.1		228.9	196.0	
	1.0	25.4	8.79	7.3	20.2		229.7	183.2	
	2.0	25.0	8.65	7.2	20.1		270.5	199.0	
	3.0	24.9	8.55	7.2	20.1		256.1	197.0	
	4.0	24.9	8.43	7.1	20.2		230.1	180.7	
	5.0	24.8	8.40	7.1	20.2		228.6	180.8	
	5.5	24.4	8.36	5.5	20.1		230.1	160.4	F2
Near Bottom	5.5	24.5	8.36	5.1	20.2		230.3	155.3	F2
UP CAST	5.0	24.8	8.43	6.8	20.1		230.8	163.4	
	4.0	25.0	8.43	7.0	20.1		231.7	177.3	
	3.0	25.1	8.61	7.0	20.0		240.3	190.4	
	2.0	25.2	8.68	7.1	20.1		227.6	180.7	
	1.0	25.5	8.82	7.2	20.1		230.1	190.9	
	0.5	25.4	8.83	7.2	20.2		229.8	199.1	
	0.1	25.5	8.83	7.4	20.2		237.6	209.4	

4981512022

04/01/2010 NCCA Field Measurement

Figure 5-2. Example Field Measurement Form (Back).

5.1.1.3 Secchi Disk

The Secchi disk is used to give a measurement of the transparency of the water column, also called the Secchi depth. This measurement is made at every station and is recorded on the datasheet. A 20 cm black and white Secchi disk is held by a non-stretch line that is marked in two tenths of a meter intervals.

Table 5-2 presents step-by-step procedures for measuring water column transparency. If the range of measurements for the three sets of depth readings is greater than 0.5 m, the entire process should be performed again. No sunglasses or any other devices should be used to shade the eyes while this procedure is being performed. The Secchi depth should be determined from the shady side of the boat during daylight hours.

Table 5-2. Sampling Procedure—Secchi Disk.

1. Remove sunglasses, hats or other devices used to shade the eyes.
2. Slowly lower the Secchi disk on the shady side of the boat until it is no longer visible and note the depth using the markings on the line (interpolate between markings to the nearest 0.1 meter).

   If the disk hits the bottom, meaning the Secchi depth is greater than the water depth, note this on the datasheet by marking the "Yes" circle under "Clear to Bottom?" and recording the depth of the water in both the "disappears" and "reappears" boxes.
3. Slowly raise the Secchi disk until it just becomes visible and note the depth.
4. Perform steps 1 and 2 two more times, noting both disappearance and reappearance depths each time.
5. Record all six measurements on the Field Measurement Form.
6. Flag any measurements that the team feels needs further comment or when a measurement cannot be made.
7. Repeat the entire process if the three sets of measurements vary by more than 0.5 m.

5.1.1.4 Multi-Probe Sonde

Dissolved Oxygen Meter

Calibrate the DO meter prior to each sampling event and record the calibration values on the Field Measurement Form. It is recommended that the probe be calibrated in the field against an atmospheric standard (ambient air saturated with water) prior to launching the boat. In addition, manufacturers typically recommend periodic comparisons with a DO chemical analysis procedure (e.g., Winkler titration) to check accuracy and linearity.

pH Meter

Calibrate the pH meter prior to each sampling event and record the calibration values on the field measurement form. Calibrate the meter in accordance with the manufacturer's instructions and with the team agency's existing Standard Operating Procedure (SOP). You must also conduct a quality control check with the provided standard to verify the calibration and periodically evaluate instrument precision (see Section 3.1.2). Once a week, each crew must check their multi-probe against the QCS provided in each site kit. Any irregularities must be reported to either the Site Kit Coordinator or the Field Logistics Coordinator immediately, and a new QCS requested. QCS values are also recorded on the Field Measurement Form.

After all *in situ* measurements have been completed for the sampling day, the team must perform a post-measurement calibration check of the pH meter to detect any drift from the values. To do this, measure the pH of the calibration standard that was used earlier in the day to calibrate the instrument. Record these values on the Field Measurement Form. If significant drift is detected, it may indicate that the meter is in need of service.

Temperature Meter

Check the accuracy of the sensor against a thermometer that is traceable to the National Institute of Standards (NIST) at least once per sampling season. The entire temperature range encountered in the NCCA should be incorporated in the testing procedure and a record of test results kept on file.

Salinity/Conductivity Meter

Calibrate the salinity/conductivity meter prior to each sampling event and record the calibration values on the field measurement form. Calibrate the meter in accordance with the manufacturer's instructions. The entire salinity/conductivity range encountered in the NCCA should be incorporated in the testing procedure and a record of test results kept on file. You must also conduct a quality control check with the provided standard to verify the calibration and periodically evaluate instrument precision (see Section 3.1.2). Once a week, each crew must check their multi-probe against the QCS that was supplied in each base kit. Any irregularities must be reported to the Field logistics coordinator immediately. QCS values are also recorded on the field measurement form.

After all *in situ* measurements have been completed for the sampling day, the team must perform a post-measurement calibration check of the conductivity meter to detect any drift from the values. To do this, measure the conductivity of the calibration standard that was used earlier in the day to calibrate the instrument. Record these values on the Field Measurement Form. If significant drift is detected, it may indicate that the meter is in need of service.

Table 5-3 presents step-by-step procedures for measuring dissolved oxygen, pH, temperature, and salinity (marine sites) or conductivity (freshwater sites).

Table 5-3. Sampling Procedure—Temperature, pH, Dissolved Oxygen and Salinity/Conductivity

1. Check meter and probes and calibrate according to manufacturer's specifications and record the calibration values on the field measurement form.
2. Check the calibration against the provided QCS solution for pH and salinity/conductivity and record the results on the field form as the QCS Measured value. This should be done at least once a week.
3. Record the true value of the QCS solution from the stock solution container on the field form as QCS True.
4. Measurements are taken at the X site at multiple depths. Measurements of all four parameters will be recorded at each depth.
5. Measure the total water depth to the nearest 0.1 meters and record on the hydrographic profile form.
6. Lower the sonde in the water and measure DO, pH, temperature, and salinity/conductivity at the following depths: 0.1 m below the surface, 0.5 m below the surface, every 1 meter from depths of 1.0 to 10.0 meters, and every 5 meters thereafter if the site is greater than 10 m. Take the last set of measurements at 0.5 m from the bottom.
7. Repeat the full sets of measurements at each of the same depth intervals as the probe is retrieved.
8. The first set of measurements will be placed in the "downcast" section of the data sheet. Take measurements at the same depth intervals on the return cast to the surface and record in the "upcast" section. Two examples are provided below that illustrate the depths at which measurements will be taken.

EXAMPLE 1:	**EXAMPLE 2:**
Water Depth = 7.2 meters	*Water Depth = 23.9 meters*
0.1 m	0.1 m
0.5 m	0.5 m
1.0 m	1.0 m
2.0 m	2.0 m
3.0 m	3.0 m
4.0 m	4.0 m
5.0 m	5.0 m
6.0 m	6.0 m
6.7 m	7.0 m
	8.0 m
	9.0 m
	10.0 m
	15.0 m
	20.0 m
	23.4 m

9. Record the measurements on the Field Measurement Form.
10. Flag any measurements that the team feels needs further comment or when a measurement cannot be made.
11. After all in situ measurements have been completed for the sampling day, perform a post-measurement calibration check of the pH and conductivity probes. Record these values on the Field Measurement Form.

5.1.1.5 Photosynthetically Active Radiation (PAR) Meter

A PAR meter, attached to a data logger such as a LI-COR®, is used to obtain a vertical profile of light in order to calculate the light attenuation coefficient at each station. PAR sensors require no field calibration, however, they should be returned to the manufacturer at least every 2 years for calibration. PAR measurements are taken at the same depths as other water column indicators. Table 5-4 presents step-by-step procedures for measuring light attenuation.

Table 5-4. Sampling Procedure—Light Attenuation (LI-1400 Datalogger).

1. Connect a deck sensor and an underwater sensor to the PAR meter. Make sure the correct calibration factors are entered for each probe. These are supplied by the manufacturer and are specific to each individual probe.
2. Place the deck sensor on the boat in a location where it is not shaded.
3. Turn on the LI-1400 meter.
4. Press the View key.
5. Using the left or right keys navigate to "NEW DATA" and press Enter.
6. Using the left or right keys navigate until channel I1I is displayed; this shows the instantaneous reading for that channel. Scrolling down will allow viewing of 2 channels at once.
7. Lower the underwater sensor on the SUNNY (or at least unshaded) side of the boat to a depth of 10 cm (represents "surface").
8. When all sensors are in place at the desired depths, it is important to record both readings at the same instant to ensure there is no change in light availability. One option is to use the datalogger by pressing ENTER and manually storing the instantaneous readings from all attached sensors.
9. To review the saved data, press Esc then use the right or left key to select "LOG DATA" press Enter.
10. Select "View=ALL", press Enter.
11. Use the down key to scroll through stored data by date and time to find the data that was just logged; press Enter to access logged data. Use the down key to view both of the sensor readings.
12. Record the values from both sensors ($\mu E/m^2/s$), along with the water depth of the underwater sensor, on the datasheet. Record the deck sensor reading in the ambient (AMB) column, and the underwater sensor reading in the underwater (UW) column.
13. Lower the underwater sensor to 1.0 meter, repeat steps 8 through 11 to capture the values, and record the values from both sensors, along with the depth of the underwater probe.
14. Repeat measurements at the following schedule *(same as other water quality measurements):*
    - 0.5 meters
    - Every 1 meter from 1.0 m to 10.0 m
    - Every 5 meters thereafter for sites greater than 10 meters
    - 0.5 meters from the bottom
15. If the bottom is impacted with the meter, allow 2-3 minutes for the disturbed conditions to settle before taking the reading.
16. If the light measurements become negative before reaching the bottom measurement, terminate the profile at that depth.
17. Repeat the process on the upcast and record values on the datasheet.

Note: Pressing the On/Off key will only turn off the screen. To shutdown the LI-1400 press the Fct key and use the right or left keys to navigate to "SHUTDOWN". Press Enter to shutdown.

5.2 Fecal Indicator (Enteroccoci)

The collection time of the Enterococci sample may vary based on whether the team will be collecting fish for fish tissue samples and whether those collections will be performed using an active or passive fishing method. Refer to section 2.1 and Figures 2-1, 2-2, and 2-3 for more information. In short, if the team is not fishing, or is using a passive fishing method, the Enterococci collection should take place immediately following the hydrographic profile. If the team is using active fishing methods, the collection of the Enterococci sample should take place at the end of the sampling day. This is based on the need to protect the Enterococci sample from potential contamination and to minimize holding times once the sample is collected. There is a strict 6 hour window from the time of collection of the Enterococci sample to the time when four aliquots of sample are filtered and all four filters frozen.

5.2.1 Summary of Method

Collect a fecal indicator sample at the X-site. Use a pre-sterilized, 250 mL bottle and collect the sample at about 0.5 meters below the water surface. For smaller vessels, this can be accomplished with a gloved hand. For larger vessels, the bottle may need to be affixed to a pole dipper. Following collection, add a sodium thiosulfate tablet, shake well, place the sample in a cooler, chill for at least 15 minutes, and maintain on ice prior to filtration of four 50 mL volumes. (Samples must be filtered and frozen on dry ice within 6 hours of collection). In addition to collecting the sample, look for signs of disturbance throughout the reach that would contribute to the presence of fecal contamination to the waterbody. Record these disturbances on the Site Assessment Form (Figure 8-2).

5.2.2 Equipment and Supplies

Table 5-5 provides the equipment and supplies needed to collect the fecal indicator sample. Record the sample data on the Sample Collection Form (Figure 5-4).

Table 5-5. Equipment and Supplies List for Fecal Indicator Sampling

For collecting samples	▪ nitrile gloves ▪ pre-sterilized, 250 mL sample bottle ▪ sodium thiosulfate tablet ▪ Wet ice ▪ cooler
For recording measurements	▪ Sample Collection Form ▪ Fecal Indicator sample labels (4 vial labels and 1 bag label) ▪ Pencils (for data forms) ▪ Fine tipped indelible markers (for labels) ▪ Clear tape strips

5.2.3 Sampling Procedure

The procedure for collecting the fecal indicator sample is presented in Table 5-6.

Table 5-6. Procedure for Fecal Indicator (Enterococci) Sample Collection

1. Put on nitrile gloves.
2. Collect the sample over the side of the boat with a gloved hand or pole dipper (on larger vessels).
3. Lower the un-capped, inverted 250 mL sample bottle to a depth of 0.5 meters below the water surface, avoiding surface scum, vegetation, and substrates. Point the mouth of the container away from the boat. Right the bottle and raise it through the water column, allowing bottle to fill completely.
4. After removing the container from the water, discard a small portion of the sample to allow for proper mixing before analyses.
5. Add the sodium thiosulfate tablet, cap, and shake bottle 25 times.
6. Store the sample in a cooler on wet ice to chill (not freeze). Chill for at least 15 minutes and do not hold samples longer than 6 hours before filtration and freezing.

5.3 Water Chemistry Sample Collection and Preservation

5.3.1 Summary of Method

The water chemistry samples will be analyzed for chlorophyll-*a*, dissolved ammonia, nitrites, nitrates, and phosphorus. Collect the water samples with either a pumped system or a water sampling bottle such as a Niskin, Van Dorn, or Kemmerer bottle and transferred to a rinsed 250 mL amber Nalgene bottle. Water for the chlorophyll-*a* sample will be collected and transferred to a separate 2 L amber Nalgene bottle. Store all samples in darkness on ice in a closed cooler. After you filter the chlorophyll-*a* samples, the filters must be kept frozen until ready to ship. A portion of the filtrate from the chlorophyll-*a* processing will be collected for the dissolved nutrient sample. Note that the fecal indicator sample IS NOT collected with these samples.

Collect the samples at the X-site, 0.5 meters below the surface.

5.3.2 Equipment and Supplies

Table 5-7 provides the equipment and supplies needed to collect water samples at the X-site.

Table 5-7. Equipment and Supplies—Water Chemistry and Chlorophyll-*a* Sample Collection

For collecting samples	▪ Water sampling device or water pumping system ▪ Nitrile gloves ▪ one 250 mL amber Nalgene bottle ▪ one 2 L amber Nalgene bottle (chlorophyll-*a*) ▪ Cooler with wet ice ▪ Field Operations Manual and/or laminated Quick Reference Guide
For recording measurements	▪ Sample Collection Form ▪ Field Measurement Form ▪ Pencils (for data forms) ▪ Fine-tipped indelible markers (for labels) ▪ Clear tape strips

5.3.3 Sampling Procedure

Table 5-8 describes the sampling procedures for collecting water chemistry samples.

Table 5-8. Sampling Procedure for Water Chemistry and Chlorophyll-*a* Sample Collection

1. Collect the water chemistry samples at the X-site (located via GPS).
2. Put on nitrile gloves. Make sure not to apply sunscreen or other chemical contaminants until after the sample is collected.
3. Using either a pumped system or a water sampling bottle, collect a water sample at 0.5 m below the surface.
4. Rinse the pumped system or sampling bottle as well as the sample containers three times with water from the site. Fill the 250 mL amber Nalgene bottle (for water chemistry) and the 2 L amber Nalgene bottle (for chlorophyll-*a*) with sample water, and place in a cooler on ice. Make sure the water chemistry label is complete and taped over and place samples in a cooler on ice at 4°C.
5. Record the collection data on the Sample Collection Form. Note anything that could influence sample chemistry (heavy rain, potential contaminants) in the Comments section. If the samples were not taken at the X-site, enter the GPS coordinates of the sampling location and the reason for relocation in the comments field on the Sample Collection Form (Figure 5-4).

5.4 Phytoplankton

5.4.1 Summary of Method

In all Great Lakes sites, teams will collect a sample for Phytoplankton analysis. Collect this sample from the X-site at the same time and depth as the other water samples. Fill a 1 liter amber nalgene bottle with from the water sampler or pumped system. The phytoplankton sample must be preserved with 10 mL of Lugol's solution within 2 hours of collection. Store the samples in darkness inside a cooler with ice or in a refrigerator.

5.4.2 Equipment and Supplies

Table 5-9 provides the equipment and supplies needed to collect the phytoplankton sample at the X-site.

Table 5-9. Equipment and Supplies—Phytoplankton

For collecting and preserving samples	▪ Water sampling device or water pumping system ▪ Nitrile gloves ▪ one 1L amber Nalgene bottle ▪ Cooler with wet ice ▪ Field Operations Manual and/or laminated Quick Reference Guide ▪ Lugol's solution
For recording measurements	▪ Sample Collection Form ▪ Pencils (for data forms) ▪ Fine-tipped indelible markers (for labels) ▪ Clear tape strips

5.4.3 Sampling Procedure

Table 5-10 describes the sampling and preservation procedures for phytoplankton samples.

Table 5-10. Sampling and Preservation Procedure for Phytoplankton

1. Collect the water chemistry samples at the X-site along with the other water samples.
2. Put on nitrile gloves. Make sure not to apply sunscreen or other chemical contaminants until after the sample is collected.
3. Using either a pumped system or a water sampling bottle, collect a water sample at 0.5 m below the surface.
4. Fill the 1 L amber Nalgene bottle with sample water, and place in a cooler on ice. Make sure the phytoplankton label is complete and taped over and place samples in a cooler on ice at 4°C.
5. The sample must be preserved by adding 10 mL of Lugol's solution to the bottle within 2 hours of collection. Rotate the bottle gently to mix the sample.
6. Record the collection data on the Sample Collection Form. Include the depth of collection, time of collection, and time of preservation.

5.5 Underwater Video

At all Great Lakes sites, a 1 minute video image of the substrate will be collected using an underwater video camera system. The video will be enhanced and examined in the lab to visually document the bottom composition, and record the presence or absence of zebra mussels, Cladophora, or other organisms.

5.5.1 Summary of Method

High quality underwater video will be best achieved if the field crew deploys the camera and records the video at approximately the same time as the *in situ* measurements and water collection activities. Avoid heavy disturbance of the bottom with anchors or sediment samplers before capturing the video images.

While anchored at the X-site, lower the camera into the water on the windward side of the boat. One person is needed to operate the DVR and one to lower the camera. The person operating the DVR should instruct the camera person on descent speed and depth of camera. When a clear view of the bottom can be seen, start recording the video by pressing the record button. Continue recording until you have captured 1 min of good bottom footage. In low light conditions, turn on the camera light by pressing the red button on the DVR end of camera cable. Experiment with the light while monitoring the screen for best picture results.

5.5.2 Equipment and Supplies

Table 5-11 provides the equipment and supplies needed to record the underwater video at the X-site.

Table 5-11. Equipment and Supplies—Underwater Video

For recording underwater video	▪ Seaviewer underwater camera ▪ Seaviewer digital video recorder (DVR) ▪ Seaviewer SeaTrak GPS overlay ▪ Garmin Etrex GPS ▪ Camera cable (100') ▪ Cable from GPS overlay to DVR ▪ Cable from GPS overlay to GPS ▪ 12v 18ah battery ▪ Charger for 12v battery ▪ Power cord (DVR ,Camera ,GPS overlay) ▪ Power adapters (110VAC – 12VDC) (3) for camera, DVR, and GPS overlay ▪ 18 gb SD card ▪ Stop watch (or similar time keeping device) ▪ Seaviewer case (all components will fit into case for transport) ▪ 10 amp fuses (Automotive blade (large) type) ▪ AA batteries (for GPS)

5.5.3 Initial Setup of Underwater Camera System

Underwater camera systems will be assembled and set up prior to shipment to field crews. However, information contained within this section will allow a field crew to verify equipment setup or troubleshoot potential connection problems. The underwater camera system and cables should be set up as shown in Figure 5-3. The system should not be disassembled between sites other than to remove the battery clips. Initial one-time setup of the camera system GPS unit is described in Table 5-12.

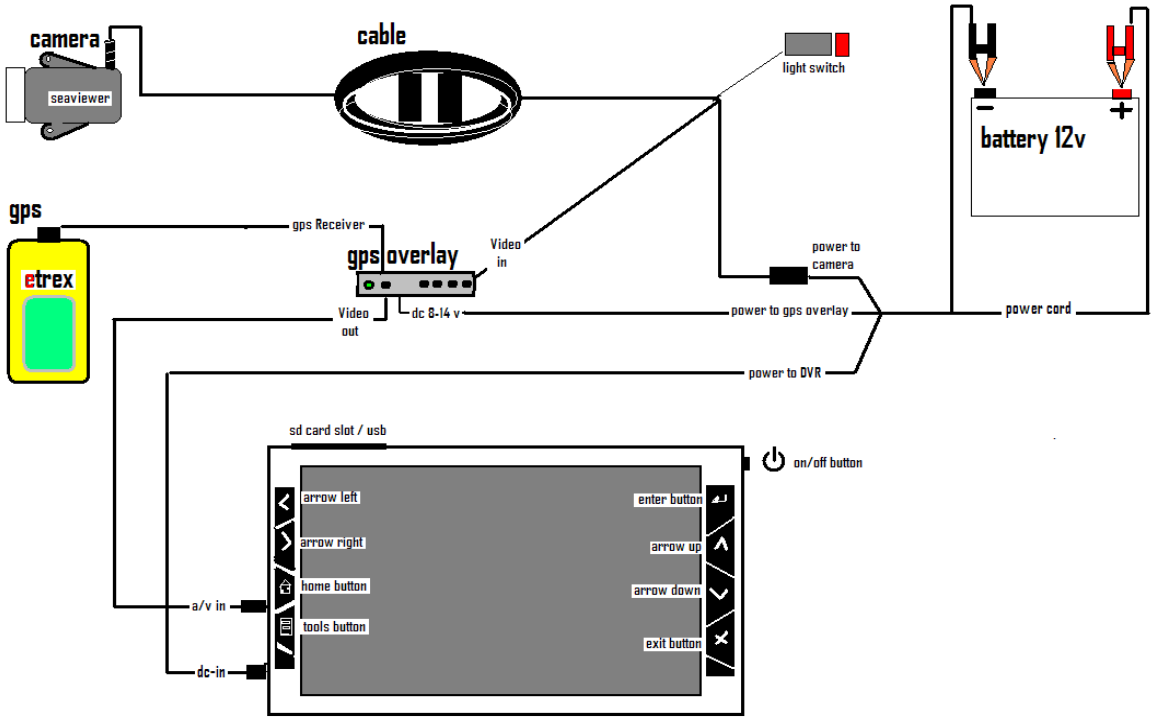


Figure 5-3. Setup Diagram of Underwater Video System

Table 5-12. Initial Setup of Camera System GPS

Set the GPS to output NMEA data in the GPS Menu section of the settings to send position information to the GPS overlay system. (This step will be completed prior to shipment of the system, but the steps below can be completed to verify correct setup).
1. Press “page” button 4 times to reach menu page.
2. Select “set up” by pressing arrow down button until “set up” is highlighted, then press enter.
3. Select “interface” by pressing arrow down button until “interface” is highlighted, then press enter.
4. Press enter again, select “NMEA out” by pressing arrow down button until “NMEA out” is highlighted, then press enter.
5. Press page button 3 times to return to satellite tracking page.

5.5.4 Underwater Video Recording Procedure

Table 5-13 describes the procedure for recording the underwater video at the X-site. Table 5-14 describes the procedure for archiving the video file after the recording has been completed.

Table 5-13. Procedure for Recording Underwater Video

1. Power up the GPS and wait until it displays: “ready to navigate” before proceeding.
2. If the GPS is not set to output NMEA data, see Table 5-12
3. Send power to the camera, GPS overlay and DVR by attaching alligator clips to the 12v battery. Attach red clip to the positive (+) terminal and black clip to the negative (-) terminal.
4. Turn on the GPS overlay by pressing its power button. The green light will illuminate.
5. Turn on the DVR by pressing the power button (upper right side of DVR) for 3 seconds. A flash screen will appear for a short time then disappear. NOTE: DO NOT PRESS POWER ON AGAIN DURING THIS TIME! A windows type menu screen will appear.
6. Initialize the DVR by pressing the video button located at top right of the DVR unit (not on the screen). A video image from the camera should appear on the DVR screen with a latitude / longitude display. (If not, see the trouble shooting section supplied with manufacturer’s literature).
7. Lower camera into the water on the windward side of the boat. One person is needed to operate the DVR and one to lower the camera. The person operating the DVR should instruct the camera person on descent speed and depth of camera.
8. When a clear image of the bottom is displayed, hold the camera as still as possible and start recording by pressing the record button (located at top right of the unit, to the right of the video button). The word “Recording” appears on the screen in red for 10 sec, then disappears. (To pause recording, press the enter button then press again to resume recording.)
9. Continue recording until you have captured 1 minute of good bottom footage.
10. In low light conditions, turn on the camera light by pressing the red button on the DVR end of camera cable. Experiment with the light while monitoring the screen for best picture results.
11. Stop recording by pressing the video key (top of unit) , or the X button.

Table 5-14. Procedure for Archiving Underwater Video Files

Upon completing the 1 minute of underwater video, it is important to verify that the video has been saved, record the file name on the Sample Collection Form, and preview the video to ensure adequate quality.

1. Select browser by pressing the enter button (upper right key on front of DVR).
2. Arrow down to "DVR".
3. Select "DVR" by pressing the enter button (upper right key front of DVR).
4. Arrow down to the last file listed. This should be the video you just recorded.
5. Record the file name on the Sample Collection Form. The format of the file is: **DVRyymmdd_hhmm_xxx.avi** (*yymmdd* is the date in year, month and day; *hhmm* is the time in hours and minutes; xxx is a file number assigned by the DVR, typically 001; and avi is the file format). Check that the date and time on the file name match the date and time of the recording you just made.
6. Press enter to play video to evaluate the quality of the video.
7. If the video clearly shows the composition of the bottom then the video is deemed acceptable; continue to step 9.
8. If the video is unviewable or is of poor quality, repeat the recording steps in Table 5-13.
9. Shut down the system by the following the steps below.
    a. Power down the DVR by pressing and holding the power button (upper right side).
    b. Power down the GPS overlay by pressing the power button.
    c. Power down the camera by disconnecting the alligator clips from the battery posts.
    d. Power down GPS by pressing and holding its power button.
10. Recharge the 12v battery at the end of each day.(It is a good idea to assign this task to an individual crew member.)
11. Back-up files to the SD card by following the steps below (do this as soon as possible to back up your data).
    a. Insert SD card into the SD slot (top left on DVR).
    b. Power on the DVR.
    c. Arrow over to "Browser" (picture of monitor with magnifying glass).
    d. Select "Browser" by pressing the enter button (upper right key on front of DVR).
    e. Arrow down to "DVR".
    f. Select "DVR" by pressing the enter button (upper right key front of DVR).
    g. Arrow down to the file you want to copy to the SD card.
    h. Press the "Tools" button (lower left key).
    h. The "CCP" option will be highlighted. Press the enter button.
    i. The "copy" option will be highlighted. Press the enter button.
    j. A second path will appear listing the SD card as an option. Arrow down to SD card.
    k. Press the enter button (upper right key front of DVR)
    l. Press the "Tools" button (lower left key).
    m. "Paste" will be highlighted. Press enter.

NOTE: backing up files to your computer is also suggested; these files are/can be large so this may not be an option for you. You can either insert the SD card directly into the computer to copy, or use the supplied USB cable to plug into your computer. Your computer will see the DVR hard drive as another drive and you can copy and paste them to a file on your computer.

NCCA 2010 SAMPLE COLLECTION FORM - (Front) Reviewed by (Initials): JPS

SITE ID: NCCA10 1009 DATE: 06/01/2010

WATER CHEMISTRY , CHLOROPHYLL and NUTRIENT COLLECTION (0.5m)

Water Chemistry (Non-Filtered)	Chilled	Comments	No Sample Collected ○
999101	●		

Chlorophyll-a	Frozen	Vol Filtered (ml)	Comments	No Sample Collected ○
999102	●	1000		

Nutrients (Filtered)	Chilled	Comments	No Sample Collected ○
999103	●		

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

ENTEROCOCCI (Target Volume = 250 mL) No Sample Collected ○

Sample ID (One unique ID per line)		Time Collected (hhmm)	Depth Collected (m)	Sample Volume (mL)	Filt. Start Time (hhmm)	Volume Filtered (Target = 250 mL) Filt. 1	Filt. 2	Filt. 3	Filt. 4	Vol. of Rinse	Filt. End Time (hhmm)	Time Frozen (hhmm)	Flag
999888		1445	0.5	250	1600	50	50	50	50	80	1730	1800	
Filter Blank						Volume Filtered (Target = 20 mL) Filt. 1	Filt. 2	Filt. 3	Filt. 4				
999889	● F					20	20	20	20				

Flag codes: K = No measurement made, U = Suspect measurement, F1,F2, etc. = flags assigned by each field crew. Explain all flags in comments.

Flag	Comments

GREAT LAKES ONLY

PHYTOPLANKTON (1-L) No Sample Collected ○

Sample ID	Preserved	Depth Collected (m)	Time Collected (hhmm)	Time Preserved (hhmm)	Comments
999100	●	0.5	1045	1130	

Under water video camera Digital Video Recording No Sample Collected ○

File name Format: DVRyymmdd_hhmm_xxx.avi		Transferred to SD Card	Comments
DVR 100601_1030_001	.avi	●	

8935052620

04/22/2010 NCCA Sample Collection

Figure 5-4. Example Sample Collection Form (Front)

6.0 SEDIMENT COLLECTIONS

6.1 Sediment Collections

Sediments are collected for a variety of analyses. One sample is collected for benthic species composition and abundance, and additional sediment grabs are collected for chemical analyses (organics/metals and TOC), grain size determination, and for use in acute whole sediment toxicity tests. The number of grabs needed may vary based on the sediment characteristics. While the biology grab is being processed (sieved), grabs should be collected for chemistry, grain size and toxicity tests. These grabs will be composited, mixed and split into four separate sample containers. A minimum of 4L of sediment will be required for the chemistry and toxicity samples.

6.1.1 Summary of Method

A 1/25 (0.04) m^2, stainless steel, Young-modified Van Veen Grab (or similar) sampler is appropriate for collecting sediment samples for both biological and chemical analyses. The top of the sampler is either hinged or otherwise removable so the top layer of sediment can be easily removed for chemical and toxicity sample collection. This gear is relatively easy to operate and requires little specialized training. For sampling in the Great Lakes, a standard Ponar grab (box size 22.9 cm x 22.9 cm with depth of 9 cm) with removable top screens may be more appropriate (USEPA 2001). Record the dimensions and sample area of the grab used on the Sample Collection Form, side 2. The area of sediment the grab collects is important for data analysis. If the grab sampler size is less than 0.03 m^2, take two grabs for the benthic macroinvertebrate collections and composite the sediment into the sieve.

6.1.2 Equipment and Supplies

Table 6-1 provides the equipment and supplies needed to collect sediment samples at the X-site. Record the Sediment Sample Collection and Preservation data on the Sample Collection Form.

Table 6-1. Equipment and Supplies—Sediment Collection

For collecting samples	▪ Young-modified Van Veen (or Ponar) grab with grab stand ▪ Weights and pads for grab ▪ Nitrile gloves ▪ Tub or bucket ▪ 0.5 mm stainless steel sieve (1.0 mm for CA, OR, WA) ▪ Sieve box or bucket ▪ Electrical tape ▪ Fine-tipped forceps ▪ Wide-mouthed funnel ▪ Alconox ▪ Formalin ▪ Rose Bengal Stain	▪ Borax ▪ Centimeter ruler ▪ Squirt bottle ▪ Stainless mixing pot or bowl with lid ▪ Cooler with wet ice ▪ Stainless steal or Teflon spoons ▪ 1 L Nalgene bottle(s) for benthos ▪ 1 gallon plastic bucket for toxicity ▪ 500 mL glass jar for organics and metals ▪ 125 mL Nalgene jar for grain size ▪ 60 mL Nalgene jar for TOC ▪ Field Operations Manual and/or laminated Quick Reference Guide
For recording measurements	▪ Sample Collection Form ▪ Field Measurement Form ▪ Pencils (for data forms) ▪ fine-tipped indelible markers (for labels) ▪ Clear tape strips	

6.1.3 Sampling Procedure

Table 6-2 describes the sampling procedure to obtain sediment samples.

Table 6-2. Sampling Procedure for Sediment Collection

1. The sampler must be thoroughly washed with Alconox prior to use at a station, then rinsed with ambient seawater/fresh or lake water to ensure no sediments remain from the previous station.
2. Attach the sampler to the end of the winch cable with a shackle and tighten the pin.
3. Cock the grab.
4. Lower the grab sampler through the water column such that travel through the last 5 meters is no faster than about 1 m/sec. This minimizes the effects of bow wave disturbance to surficial sediments.
5. Allow a moment for the sampler to settle into the substrate and then slack off on the cable. Letting the cable go slack serves to release the jaws of the sampler so they will close as the sampler is retrieved.
6. Retrieve the sampler and lower it into its cradle on-board. Open the top and determine whether the sampling is successful or not. A successful grab is one having relatively level, intact sediment over the entire area of the grab, and a sediment depth at the center of at least 7 centimeters for the benthic macroinvertebrate grab (see Figure 6-1). Grabs containing no sediment, partially filled grabs, or grabs with shelly substrates or grossly slumped surfaces are unacceptable. Grabs completely filled to the top, where the sediment is in direct contact with the hinged top, are also unacceptable. It may take several attempts using different amounts of weight to obtain the first acceptable sample. The more weight added, the deeper the bite of the grab. In very soft mud, pads may be needed to prevent the sampler from sinking in the mud. If pads are used, the rate of descent near the bottom should be slowed even further to reduce the bow wave.
7. If, after several attempts, only grabs less than 7 centimeters deep can be obtained, use the next successful grab regardless of the depth of sediment at the center of the grab. Flag the collection on the data form and be sure to accurately record the depth of the grab.
8. Carefully drain overlying water from the grab. If the grab is used for benthic community analysis, the water must be drained into the container that will receive the sediment to ensure no organisms are lost.
9. Enter notes on the condition of the sample (smell, substrate, presence of organisms on the surface, etc.) on the data sheet (Figure 6-2).
10. If the grab sampler size is less than 0.03 m^2, take two grabs for the benthic macroinvertebrate collections and composite the sediment into the sieve.
11. Process the grab sample for either benthic community analysis or chemistry/toxicity testing as described in and in Sections 6.2 and 6.3.
12. Repeat steps 4-8 until all samples are successfully collected. To minimize the chance of sampling the exact same location twice, the boat engines can be turned periodically to change the drift of the boat, or additional anchor line can be let out.

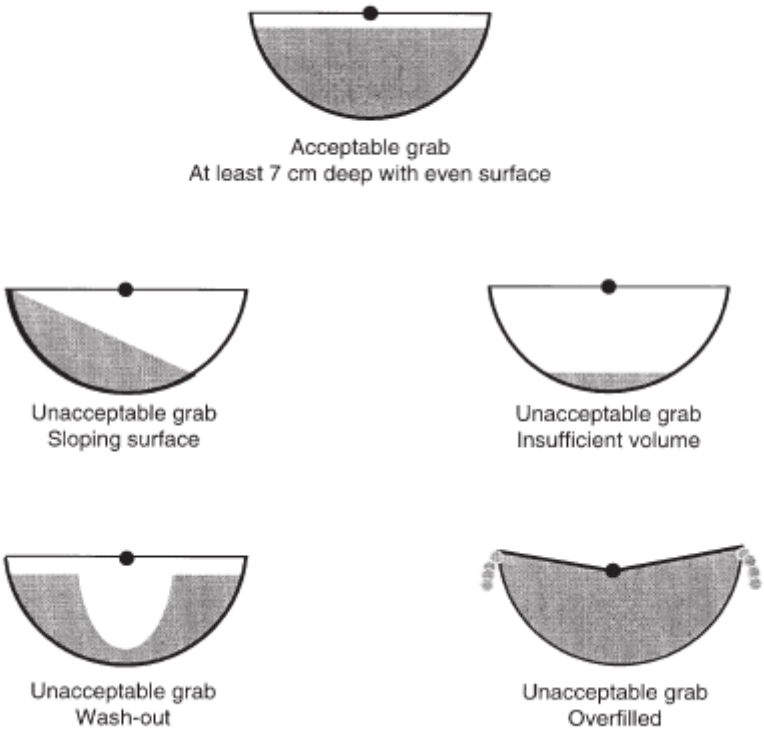


Figure 6-1. Illustration of Acceptable and Unacceptable Grabs for Benthic Community Analysis. *An acceptable grab is at least 7 cm in depth (using a 0.04m^2 Van Veen sampler), but not oozing out of the top of the grab, and has a relatively level surface.*

6.2 Benthic Macroinvertebrate Composition And Abundance

6.2.1 Field Processing of Benthic Macroinvertebrate Samples

Grab samples obtained to assess the benthic macroinvertebrate community are processed as outlined in Table 6-3.

Table 6-3. Processing Procedure for Benthic Macroinvertebrate Samples

1. Measure the depth of the sediment at the middle of the sampler and record the value on the data sheet. The depth should be ≥7 cm. Record descriptive information about the grab, such as the presence or absence of a surface floc, color and smell of surface sediments, and visible fauna on the data sheet.

2. Dump the sediment into a clean basin and then into a 0.5 mm mesh sieve (1.0 mm mesh in CA, OR, WA). Place the sieve into a table (sieve box) containing water from the sampling station, a larger bucket or place over the side of the boat. Agitate the tray in the sieve box thus washing away sediments and leaving organisms, detritus, sand particles, and pebbles larger than 0.5 mm or 1.0 mm where relevant. This method minimizes mechanical damage to fauna that is common when forceful jets of water are used to break up sediments. A gentle flow of water over the sample is acceptable. Extreme care must be taken to assure that no sample is lost over the side of the sieve.

3. Drain the water from the sieve box and gently rinse the contents of the tray to one edge. Remove large non-living items such as rocks and sticks after inspecting them and ensuring that **all** benthic organisms are included in the collection. Using either your fingers or a spoon, GENTLY scoop up the bulk of the sample and place it in the 1 L Nalgene screw-top bottle (which should be placed in the sieve or a bucket in case some of the sample spills over).

4. Rinse the outside of the sample jar into the sieve, then, using a funnel, rinse the contents into the jar. The jar should be filled no more than one-half full. If the quantity of sample exceeds 500 mL, place the remainder of the sample in a second container with a "2 of 2" label. For samples with a large amount of benthos, additional jars may be needed.

5. Be sure to place the sample and jar number on a waterproof label and place inside the sample container. Record the total number of jars on the Sample Collection Form (Figure 6-2).

6. Carefully inspect the sieve to ensure that all organisms are removed. Use fine forceps (if necessary) to transfer fauna from the sieve to the bottle containing the proper sample number.

7. 100% percent buffered formalin is used to fix and preserve benthic samples. A 100 % buffered, stained stock formalin solution should be mixed according to the directions in Table 3-2. 100 mL of the formalin should be added to each sample jar along with an additional teaspoon-full of borax is to ensure saturation of the buffer.. FILL THE JAR TO THE RIM WITH SEAWATER TO ELIMINATE ANY AIR SPACE. This eliminates the problem of organisms sticking to the cap because of sloshing during shipment. Teams may choose to use a more dilute formalin solution in larger quantities as long as the end concentration of the preservative is between 6 and 7 percent.

8. Use a pencil to fill out waterproof benthos label(s) with the pertinent sample information and place it inside the bottle(s).

9. Seal the lid with electrical tape. If the sample occupies more than one container, label all the sample bottles containing material from that grab together. Each benthos jar from a single site will have the same sample ID number. Gently rotate the bottle to mix the contents and place in the dark.

10. Prior to sieving the next sample, use copious amounts of forceful water and a stiff brush to clean the sieve, thereby minimizing cross-contamination of samples.

NCCA 2010 SAMPLE COLLECTION FORM - (Back)

Reviewed by (initial): JPS

SITE ID: NCCA10 LA0000 DATE: 06/01/2010

BENTHIC INFAUNA COLLECTION

No Sample Collected ○

GRAB AREA (m²): 0.05 GRAB TYPE: ○ Van Veen ● Standard Ponar ○ Other:

SIEVE SIZE: ● 0.5 mm ○ 1.0 mm NUMBER OF GRABS: ● 1 ○ 2 NOTE: 2 Grabs are required for samplers less than 0.03 m²

Sample ID	Depth (cm) (Must be >7 cm)	No. of Jars	Preserved	Comments
999108	12	02	●	

SEDIMENT CHARACTERISTICS (Benthic Grab)

COLOR: ○ Black ○ Brown ○ Light Brown ● Dark Brown ○ Gray ○ Other

SUBSTRATE: ○ Sand ● Muck ○ Gravel ○ Cobble ○ Shellhash ○ Other

SMELL: ○ Fishy ○ Chemical ● Sulphur ○ None ○ Other

SURFACE: ○ Film ○ Floc ● Nothing Noted ○ Other

VISIBLE FAUNA: ● Yes ○ No TYPE: OLIGOCHAETE WORMS

VISIBLE FLORA: ● Yes ○ No TYPE: MACRO ALGAE

SEDIMENT ORGANICS/METALS (Glass Jar 500 ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 250 mL)	Chilled	Comments
999104	250	●	

SEDIMENT GRAIN SIZE (Nalgene 125ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 100 mL)	Chilled	Comments
999106	100	●	

SEDIMENT TOC (Nalgene 60ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 50 mL)	Chilled	Comments
999107	50	●	

SEDIMENT TOXICITY (1 gal Screw Top Bucket)

No Sample Collected ○

Sample ID	Sample Volume (Target = 3L)	Chilled	Comments
999105	3500	●	

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

3003052621

Figure 6-2. Example Sample Collection Form (Back)

6.3 Sediment Composition, Chemistry And Toxicity

In addition to grab samples collected for benthic community analysis, additional grabs are collected for chemical analyses (organics/metals and TOC), grain size determination, and for use in acute toxicity tests. The top two centimeters of these grabs are removed, homogenized, and split into these four sample types.

6.3.1 Field Processing of Sediment Samples for Chemistry and Toxicity Testing

Because of contamination concerns, these samples are removed and processed in the order described below in Table 6-4.

Table 6-4. Processing Procedure for Sediment Composition, Chemistry and Toxicity Testing

1. As each grab is retrieved, carefully examine it to determine acceptability. The grab is considered acceptable as long as the surface layer is intact. The grab need not be greater than 7 cm in depth for chemistry samples, but the other criteria outlined above apply. Carefully drain off, or siphon, any overlying water, and remove and discard large, non-living surface items such as rocks or pieces of wood. Remove any submerged aquatic vegetation (SAV) after recording its presence on the field data sheet.

NOTE: Great care must be taken to avoid contamination of this sample from atmospheric contaminants. The boat engine should be turned off or the boat maneuvered to ensure the exhaust is down wind. All containers, including the grab sampler, should be kept closed except for when opening is necessary to remove or add samples.

2. A clean stainless steel or Teflon spoon is used to remove sediments from grab samples for these analyses. The stainless steel or Teflon spoon must be washed with Alconox and rinsed with ambient seawater or lake water before use in obtaining grab samples.

3. Remove the top two cm of sediment using the stainless steel or Teflon spoon. Sediment which is in direct contact with the sides of the sampler should be excluded as they may be contaminated from the device. Place the sediment in a stainless steel pot or bowl and place the pot in a cooler on wet ice (NOT dry ice). The sample must be stored at 4°C, NOT FROZEN.

4. Repeat obtaining sediment samples from the grab and compositing the sediment in the same stainless pot until a sufficient quantity of sediment has been collected for all samples (approximately 4L). Stir sediment homogenate after every addition to the composite to ensure adequate mixing. Keep the container covered and in the cooler between grabs.

5. Homogenize the sediment by stirring with a Teflon paddle or stainless steel spoon for 10 minutes. Divide the composite into the following four sample types.

6. ORGANICS and METALS - Using a stainless steel spoon, carefully place 250 mL of sediment in a 500 mL glass bottle for chemical analysis. CARE MUST BE TAKEN TO ENSURE THAT THE INSIDE OF THE BOTTLE, BOTTLE CAP, AND THE SAMPLE IS NOT CONTAMINATED. Record the sample number, wrap the jar in "bubble wrap" to protect it from breakage, and place the sample on wet ice (NOT dry ice). To reduce the possibility of breakage, the sample should be stored at 4°C, NOT FROZEN.

7. SEDIMENT GRAIN SIZE - Using a stainless steel spoon, place approximately 100 mL of sediment into a clean 125 mL Nalgene sampling jar. Record the sample number and keep on ice at 4°C. Store this sample on wet ice (NOT dry ice).

9. TOTAL ORGANIC CARBON - Using a stainless steel spoon, place approximately 50 mL of sediment into a pre-cleaned 60 mL Nalgene sampling jar. Record the sample number and keep on wet ice at 4°C.

10. SEDIMENT TOXICITY - Using the stainless steel spoon, fill approximately 75-85% of 1 gallon plastic bucket for toxicity testing with sediment (minimum volume required is 3000 mL). Record the sample number and place the sample on wet ice (NOT dry ice). The sample must be stored at 4°C, NOT FROZEN.

7.0 FISH TISSUE COLLECTION

Fish will be collected at all NCCA sites to be analyzed for whole body concentrations of organic and inorganic contaminants to generate data for ecological purposes ("eco" fish tissue samples). Results will be used to evaluate ecological risks associated with fish consumption by wildlife.

An additional fish tissue sample for human health contaminant analysis will be taken at 150 Great Lakes sites. Refer to section 7.2 for detailed information regarding this sample.

7.1 Ecological Contamination Fish Collection

Ecological fish tissue collection will be based on biogeographically specific "target species" lists developed for each of the regional areas- Great Lakes, Northeast, Southeast, Gulf, and West Coast. In the event that target species cannot be caught at a site, then species of similar habit/habitat may be substituted. All attempts should be made to collect the targeted species. Teams are not required to expend any more than 3 hours in attempting to collect the ecological fish tissue sample. Teams may, however, spend additional time fishing if desired.

Any reasonable method which represents the most efficient or best use of the available time on station may be used to collect the fish (e.g., otter trawl, hook and line, gill net, seine, etc.). It is not recommended that specimens be obtained by purchasing fish dockside unless it can be documented that the fish purchased came from an area in close proximity to the X-site (i.e. within 500 meters). Record pertinent information regarding the fish collection method, start and stop times, and fishing location on the Eco Fish Collection Form, side 1 (Figure 7-1).

Specimens collected should be identified to species and measured to the nearest millimeter (total length). Record the taxonomic name (genus-species) and the length of each fish on the Eco Fish Collection Form, side 2 (Figure 7-2). The minimum length for a specimen for ecological risk purposes is 100 mm with a preferred length of 100 – 400 mm. A minimum of 5 individuals should be collected for the composite sample. All individuals must be of similar size, such that the smallest individual in the composite is no less than 75% of the total length of the largest individual. Up to 20 individuals (a total of 500 g of whole body tissue is needed) should be collected and retained for analysis. If it is suspected that 20 individuals will yield less than 500 g total weight, additional specimens should be collected. The lengths of any additional fish should be recorded on a supplemental fish collection form and submitted along with the Eco Fish Collection Form.

7.1.1 Equipment and Supplies for Eco Fish Tissue Sampling.

Table 7-1 lists the equipment and supplies necessary for field crews to collect ecological fish tissue samples. This list is comparable to the checklist presented in Appendix A, which provides information to ensure that field teams bring all of the required equipment to the site. Additional Eco Fish Collection Supplies can be ordered from GLEC at 231-941-2230.

Table 7-1. Equipment and Supplies—Eco Fish Tissue Collection

For collecting fish composite sample	▪ Scientific collection permit ▪ Otter trawl (or other device to collect sufficient sample) ▪ Sampling vessel (including boat, motor, trailer, oars, gas, and all required safety equipment)	▪ Coast Guard-approved personal floatation devices ▪ Global Positioning System (GPS) unit ▪ Livewell and/or buckets ▪ Measuring board (millimeter scale) ▪ Clean nitrile gloves ▪ Wooden bat
For storing and preserving fish composite sample	▪ 2 gallon plastic self-sealing bags ▪ Large plastic (composite) bags	▪ Coolers ▪ Plastic cable ties ▪ Dry ice or wet ice (for temporary transport)
For documenting the fish composite sample	▪ Field Record Forms ▪ Clipboard ▪ #2 pencils	▪ Sample Identification Labels ▪ Tyvek label tags ▪ Fine tipped indelible markers
For shipping the fish composite samples	▪ Preaddressed FedEx airbill ▪ Coolers ▪ Dry ice (50 lbs per cooler)	▪ Tracking Form ▪ Packing/strapping tape

7.1.2 Sampling Procedure

The eco fish tissue samples will be collected using any reasonable method that is most efficient and the best use of available time on station. Fish tissue sample collection occurs in proximity to the X-site where other indicator samples are obtained (within a 500 meter radius of the X-site). Record the method(s) used on the Eco Fish Collection Form.

At least five individuals of the target species are needed, yielding a minimum of 500 g total weight. If a full composite sample is not collected after 3 hours of effort, teams may terminate the sampling and submit as many fish as possible. Record the details of the sample on the Eco Fish Collection Form. If the target species are unavailable, the fisheries biologist will select an alternative species (i.e., a species that is commonly present in the study area, with specimens of harvestable or consumable size, and in sufficient numbers to yield a composite) to obtain a fish composite sample from the species that are available. **Recommended** target species are given in Tables 7-2 and 7-3.

Table 7-2. Recommended Great Lakes Target Species for Whole Body Fish Tissue Collection by Lake

	Family name	Common name	Scientific name
Lake Erie	Cyprinidae	Common carp	*Cyprinus carpio*
	Gobiidae	Round goby	*Neogobius melanostomus*
	Ictaluridae	Channel catfish	*Ictalurus punctatus*
	Moronidae	White perch	*Morone americana*
		White bass	*Morone chrysops*
	Percidae	Yellow perch	*Perca flavescens*
		Walleye	*Sander vitreus*
	Sciaenidae	Freshwater drum	*Aplodinotus grunniens*
Lake Huron	Centrarchidae	Smallmouth bass	*Micropterus dolomieu*
	Cottidae	Slimy sculpin	*Cottus cognatus*
	Percidae	Yellow perch	*Perca flavescens*
		Walleye	*Sander vitreus*
Lake Superior	Osmeridae	American / Rainbow smelt	*Osmerus mordax*
	Salmonidae	Lake whitefish	*Coregonus clupeaformis*
		Cisco / Lake Herring	*Coregonus Artedi*
		Lake trout	*Salvelinus namaycush*
Lake Ontario	Catostomidae	Shorthead redhorse	*Moxostoma macrolepidotum*
	Centrarchidae	Rock bass	*Ambloplites rupestris*
		Pumpkinseed	*Lepomis gibbosus*
		Bluegill	*Lepomis macrochirus*
		Smallmouth bass	*Micropterus dolomieu*
		White crappie	*Pomoxis annularis*
		Black crappie	*Pomoxis nigromaculatus*
	Cottidae	Mottled sculpin	*Cottus bairdii*
		Slimy sculpin	*Cottus cognatus*
	Cyprinidae	Common carp	*Cyprinus carpio*
		Lake chub	*Couesius plumbeus*
		Bluntnose minnow	*Pimephales notatus*
	Esocidae	Northern pike	*Esox lucius*
		Muskellunge	*Esox masquinongy*
	Gasterosteidae	Three-spined stickleback	*Gasterosteus aculeatus aculeatus*
	Gobiidae	Round goby	*Neogobius melanostomus*
		Tubenose goby	*Proterorhinus marmoratus*
	Ictaluridae	Brown bullhead	*Ameiurus nebulosus*
		Stonecat	*Noturus flavus*
		Channel catfish	*Ictalurus punctatus*
	Lotidae	Burbot	*Lota lota*
	Moronidae	White perch	*Morone americana*
		White bass	*Morone chrysops*
	Percidae	Ruffe	*Gymnocephalus cernuus*
		Yellow perch	*Perca flavescens*
		Logperch	*Percina caprodes*
		Sauger	*Sander canadensis*
		Walleye	*Sander vitreus*
	Percopsidae	Trout-perch	*Percopsis omiscomaycus*
	Salmonidae	Pink salmon	*Oncorhynchus gorbuscha*
		Coho salmon	*Oncorhynchus kisutch*
		Rainbow trout	*Oncorhynchus mykiss*
		Lake whitefish	*Coregonus clupeaformis*
		Chinook salmon	*Oncorhynchus tshawytscha*
		Lake trout	*Salvelinus namaycush*
	Sciaenidae	Freshwater drum	*Aplodinotus grunniens*
Lake Michigan	Centrarchidae	Rock bass	*Ambloplites rupestris*
		Pumpkinseed	*Lepomis gibbosus*
		Bluegill	*Lepomis macrochirus*
	Cottidae	Mottled sculpin	*Cottus bairdii*
		Slimy sculpin	*Cottus cognatus*
	Cyprinidae	Lake chub	*Couesius plumbeus*
		Bluntnose minnow	*Pimephales notatus*
	Gobiidae	Round goby	*Neogobius melanostomus*
		Tubenose goby	*Proterorhinus marmoratus*
	Lotidae	Burbot	*Lota lota*
	Percidae	Yellow perch	*Perca flavescens*
		Logperch	*Percina caprodes*
	Salmonidae	Lake whitefish	*Coregonus clupeaformis*
		Lake trout	*Salvelinus namaycush*

Table 7-3. Recommended Marine Target Species for Whole Body Fish Tissue Collection by Specific Biogeographical Region

	Family name	Common name	Scientific name
Northeast	Ictaluridae	White catfish	*Ameiurus catus*
		Channel catfish	*Ictalurus punctatus*
	Moronidae	White perch	*Morone americana*
	Paralichthyidae	Summer flounder	*Paralichthys dentatus*
	Pleuronectidae	Winter flounder	*Pseudopleuronectes americanus*
	Sciaenidae	Gray weakfish	*Cynoscion regalis*
	Sparidae	Scup	*Stenotomus chrysops*
	Nephropoidea	Lobster	*Homarus americanus*
Southeast/ Gulf of Mexico	Ariidae	Hardhead sea catfish	*Ariopsis felis*
		Gafftopsail sea catfish	*Bagre marinus*
	Paralichthyidae	Southern flounder	*Paralichthys lethostigma*
		Gulf Flunder	*Paralichthys albigutta*
		Summer flounder	*Paralichthys dentatus*
	Sciaenidae	Sand weakfish (or seatrout)	*Cynoscion arenarius*
		Spot croaker	*Leiostomus xanthurus*
		Gray weakfish	*Cynoscion regalis*
		Atlantic croaker	*Micropogonias undulatus*
		Speckled Trout	*Cynoscion nebulosus*
		Red Drum	*Sciaenops ocellatus*
	Sparidae	Pinfish	*Lagodon rhomboides*
West Coast	Atherinopsidae	Topsmelt silverside	*Atherinops affinis*
	Cottidae	Pacific staghorn sculpin	*Leptocottus armatus*
		Saddleback sculpin	*Oligocottus rimensis*
	Cynoglossidae	California tonguefish	*Symphurus atricaudus*
	Embiotocidae	Shiner perch	*Cymatogaster aggregata*
		Striped sea perch	*Embiotoca lateralis*
	Gasterosteidae	Three-spined stickleback	*Gasterosteus aculeatus aculeatus*
	Paralichthyidae	Pacific sanddab	*Citharichthys sordidus*
		Speckled sanddab	*Citharichthys stigmaeus*
		California flounder	*Paralichthys californicus*
	Pleuronectidae	Butter sole	*Isopsetta isolepis*
		English sole	*Parophrys vetulus*
		Starry flounder	*Platichthys stellatus*
		Pacific sand sole	*Psettichthys melanostictus*
	Sciaenidae	White croaker	*Genyonemus lineatus*
	Serranidae	Spotted sand bass	*Paralabrax maculatofasciatus*
		Barred sand bass	*Paralabrax nebulifer*

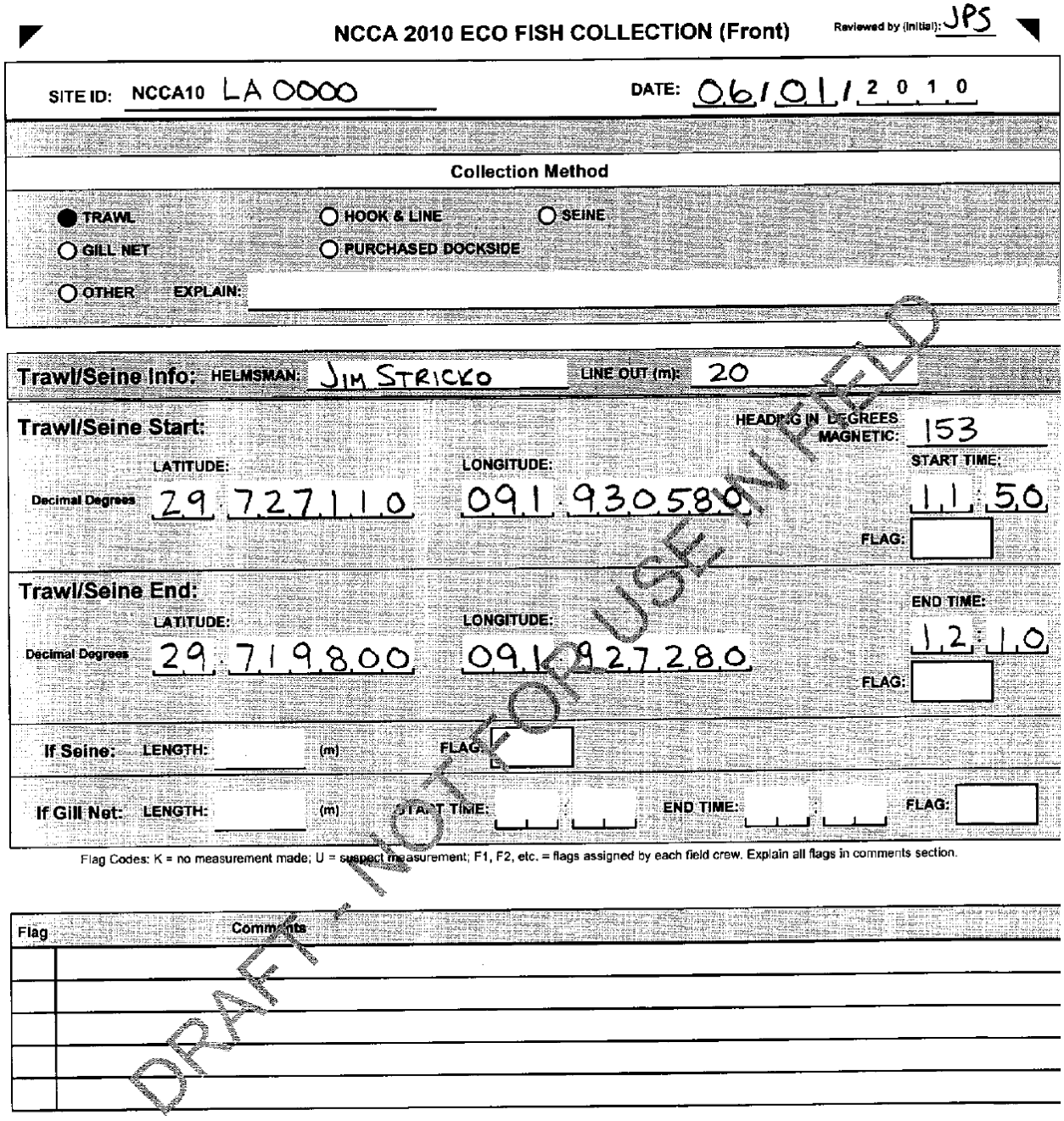
The procedures for collecting and processing fish composite samples are presented in Table 7-4.

Table 7-4. Sampling Procedure for Eco Fish Tissue Composite Samples

1. Put on clean nitrile gloves before handling the fish. Do not handle any food, drink, sunscreen, or insect repellant until after the composite sample has been collected, measured, and bagged.
2. Rinse potential target species/individuals in ambient water to remove any foreign material from the external surface and placed in clean holding containers (e.g., livewells, buckets)..
3. The eco fish composite must consist of at least 5 fish of adequate size to provide a total weight of 500 grams of whole-body tissue. Select fish for the composite based on the following criteria:
   - all are of the same species;
   - all are of similar size, so that the smallest individual in a composite is no less than 75% of the total length of the largest individual; and
   - all are collected at the same time, i.e., collected as close to the same time as possible, but no more than one week apart (Note: individual fish may have to be frozen until all fish to be included in the composite are available for delivery to the designated laboratory).
4. Identify the fish to species and record the scientific name of the Fish Tissue Data Form (Figure 7-2). Accurate taxonomic identification is essential in assuring and defining the organisms that have been composited and submitted for analysis. Individuals from different species should **not** be used in a single sample.

Table 7-4. Sampling Procedure for Eco Fish Tissue Composite Samples

5. Measure each individual fish to determine total body length. Measure total length of each specimen in millimeters, from the anterior-most part of the fish to the tip of the longest caudal finray (when the lobes of the caudal fin are depressed dorsoventrally).
6. Record collection method, sample number, species retained, specimen lengths, location collected and sampling date and time on the Fish Collection Form (Figure 7-1). Make sure the sample ID numbers recorded on the collection form match those on the sample labels.
7. Remove each fish retained for analysis from the clean holding container(s) (e.g., livewell) using clean nitrile gloves. Dispatch larger fish using a clean wooden bat (or equivalent wooden device).
8. Place each fish in a 2 gallon self-sealing bag. All fish from the composite sample should be placed in the same bag. Be careful of fish with spines that may pierce the bag. If spines are likely to puncture the bag, break off or clip the spines with a clean side-cutter or other appropriate tool and place the spine in the bag with the fish. If all the fish collected will not fit in a single 2 gallon bag, use additional bags as necessary.
9. Prepare interior and exterior Sample Identification Labels for the 2 gallon bag(s), ensuring that the label information matches the information recorded on the Fish Collection Form. **Be sure to include fish species and maximum and minimum lengths on the labels**. Place the interior label inside a small (sandwich size) self-sealing bag and place it inside the 2 gallon bag with the fish composite. Affix the exterior label to the 2 gallon bag and cover with clear plastic tape. If additional 2 gallon bags are used, fill out extra labels with the same sample ID and information for each bag and label accordingly (i.e. bag 2 of 2).
10. Double-bag the entire set of specimens in the composite, that is, place all the 2 gallon bags in the composite from the site inside a large plastic bag.
11. Prepare a Sample Identification Label for the outer bag, ensuring that the label information matches the information recorded on the Fish Collection Form. **Be sure to include fish species and maximum and minimum lengths on the label.** Affix the sample label to a Tyvek tag and cover with clear plastic tape. Thread a cable tie through the grommet in the Tyvek tag and seal the outer bag with the cable tie.
12. After the sample is packaged, place it immediately on dry ice for shipment. If samples will be carried back to a laboratory or other facility to be frozen before shipment, wet ice can be used to transport bagged fish samples in the coolers to a laboratory or other interim facility.
13. Samples may be stored on dry ice for a maximum of 24 hours. You have the option, depending on site logistics, of:
    - shipping the samples packed on sufficient quantities of dry ice (50 pounds minimum, layered to ensure direct contact between fish and dry ice) to keep samples frozen for up to 48 hours, via priority overnight delivery service (e.g., Federal Express), so that they arrive at the sample preparation laboratory within less than 24 hours from the time of sample collection, or
    - freezing the samples within 24 hours of collection at ≤-20°C, and storing the frozen samples until shipment within 2 weeks of sample collection (frozen samples will subsequently be packed on at least 50 pounds of layered dry ice and shipped to the sample preparation laboratory via priority overnight delivery service).



NCCA 2010 ECO FISH COLLECTION (Front)

Reviewed by (initial): JPS

SITE ID: NCCA10 LA 0000 DATE: 06/01/2010

Collection Method

● TRAWL ○ HOOK & LINE ○ SEINE
○ GILL NET ○ PURCHASED DOCKSIDE
○ OTHER EXPLAIN:

Trawl/Seine Info: HELMSMAN: Jim Stricko LINE OUT (m): 20

Trawl/Seine Start:

HEADING IN DEGREES MAGNETIC: 153

LATITUDE: LONGITUDE: START TIME:

Decimal Degrees 29.727110 091.930580 11:50

FLAG:

Trawl/Seine End:

LATITUDE: LONGITUDE: END TIME:

Decimal Degrees 29.719800 091.927280 12:10

FLAG:

If Seine: LENGTH: (m) FLAG:

If Gill Net: LENGTH: (m) START TIME: : END TIME: : FLAG:

Flag Codes: K = no measurement made; U = suspect measurement; F1, F2, etc. = flags assigned by each field crew. Explain all flags in comments section.

Flag	Comments

DRAFT NOT FOR USE

03/31/2010 NCCA ECO Fish Collection

0508346545

Figure 7-1. Example Eco Fish Collection Form (Front)

NCCA 2010 ECO FISH COLLECTION (Back) Reviewed by (initial): JPS

SITE ID: NCCA10 LA0000 DATE: 06/01/2010 PAGE: 1 OF 1

FISH TISSUE SAMPLE ○ NO SAMPLE COLLECTED

SAMPLE ID 999109 FISH ALL WITHIN 75% OF LARGEST SPECIMEN ● FISH ARE ALL THE SAME SPECIES ●

	Genus Species	Total Length (mm) Frozen		Comments
.1	LAGODON RHOMBOIDES	423	●	
.2		322	●	
.3		318	●	
.4		375	●	
.5		403	●	
.6			○	
.7			○	
.8			○	
.9			○	
.10			○	
.11			○	
.12			○	
.13			○	
.14			○	
.15			○	
.16			○	
.17			○	
.18			○	
.19			○	
.20			○	

FOR USE IN FIELD

The eco fish composite must consist of at least 5 fish of adequate size to provide a total weight of 500 grams of whole-body tissue.

9233044667

03/31/2010 NCCA ECO Fish Collection (Back)

Figure 7-2. Example Eco Fish Collection Form (Back)

7.2 Summary Of Method For Human Health Fish Tissue Sampling

Human health fish composites will be collected at a subset of 150 of the Great Lakes sites (30 sites per lake), and fillet tissue will be analyzed for mercury, fatty acids, and contaminants of emerging concern (e.g., perfluorinated compounds or PFCs). This section contains the sampling procedures and target species for human health fish tissue collection. Note that the human health fish species table (Table 7-6) includes 26 priority target species and 18 alternative fish species. Field crews should attempt to collect a priority target species wherever possible. If priority target species are not available at a particular site, then the field crew should collect a composite of one of the alternative fish species. In the event that a team is unable to collect fish which are on the human health species list, then the field crew should contact either Leanne Stahl, Great Lakes Human Health Fish Tissue Study Manager (U.S. EPA Office of Water), at 202-566-0404 or Blaine Snyder, Tara Kolodiej or Carolina Gallardo of Tetra Tech, Inc. at 410-356-8993 for further direction.

Any reasonable method which represents the most efficient or best use of the available time on station may be used to collect the fish (e.g., gill net, otter trawl, or hook and line). **Purchasing fish is not an option for human health fish tissue collection**. Record sample collection information on the Human Health Fish Collection Form, side 1 (Figure 7-3).

Specimens collected for each composite should be identified using scientific names (genus and species). Record the scientific name on the Human Health Fish Collection Form, side 2 (Figure 7-4), along with the total length (to the nearest mm) for each specimen in the composite. Human health fish composites should consist of 5 similarly sized (i.e., the total length of the smallest specimen is no less than 75% of the total length of the largest specimen) adult fish of the same species that will collectively yield about 500 g of fillet tissue.

Table 7-5 lists the equipment and supplies necessary for field crews to collect human health fish tissue samples. Additional Eco Fish Collection Supplies can be ordered from Tetra Tech at 410-356-8993. A list of frequently asked questions and responses will be provided with the fish sampling supplies to clarify situations that field crews may encounter while collecting human health fish composites. The procedures for collecting and processing fish composite samples are presented in Table 7-7.

Table 7-5. Equipment and Supplies—Human Health Fish Tissue Collection

For collecting fish composite sample	▪ Scientific collection permit ▪ Otter trawl (or other device to collect sufficient sample) ▪ Sampling vessel (including boat, motor, trailer, oars, gas, and safety equipment) ▪ Clean nitrile gloves	▪ Coast Guard-approved personal floatation devices ▪ Global Positioning System (GPS) ▪ Livewell and/or buckets ▪ Measuring board (millimeters) ▪ Wooden bat
For storing and preserving fish composite sample	▪ Solvent rinsed aluminum foil ▪ Food-grade poly tubing ▪ Large plastic (composite) bags	▪ Coolers ▪ Plastic cable ties ▪ Dry ice or wet ice (for temporary transport)
For documenting the fish composite sample	▪ Field Record Forms ▪ Clipboard ▪ #2 pencils	▪ Sample Identification Labels ▪ Tyvek label tags ▪ Fine tipped indelible markers
For shipping the fish composite samples	▪ Preaddressed FedEx airbill ▪ Coolers ▪ Dry ice (50 lbs per cooler)	▪ Tracking Form ▪ Packing/strapping tape

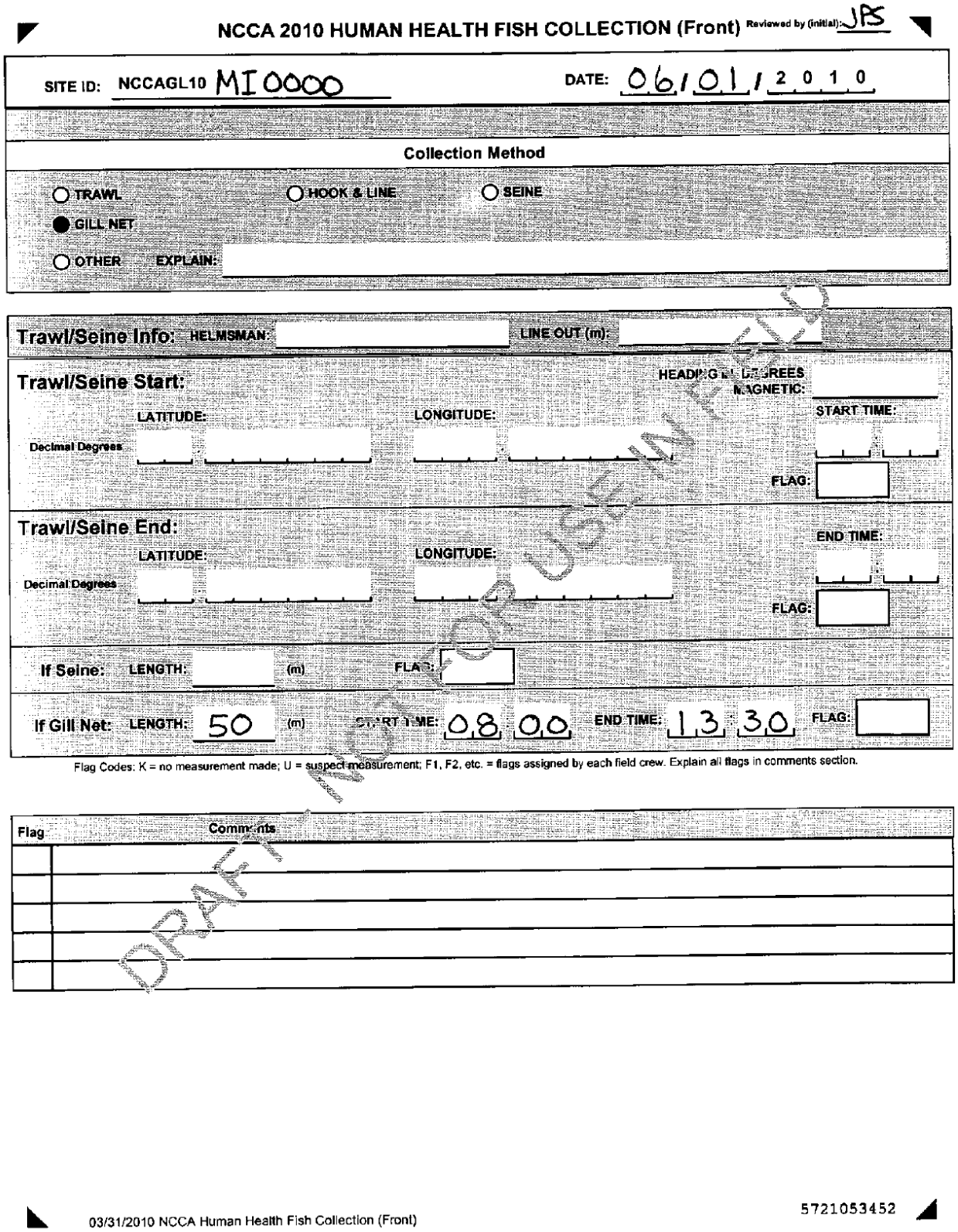
Table 7-6 Target Fish Species for Great Lakes Human Health Fish Tissue Study Fillet Composites

Priority Target Fish Species		
Family Name	**Common Name**	**Scientific Name**
Centrarchidae	Rock bass	*Ambloplites rupestris*
	Smallmouth bass	*Micropterus dolomieu*
	Largemouth bass	*Micropterus salmoides*
	White crappie	*Pomoxis annularis*
	Black crappie	*Pomoxis nigromaculatus*
Cyprinidae	Common carp	*Cyprinus carpio*
Esocidae	Northern pike	*Esox lucius*
	Muskellunge	*Esox masquinongy*
	Chain pickerel	*Esox niger*
Ictaluridae	Channel catfish	*Ictalurus punctatus*
Lotidae	Burbot	*Lota lota*
Moronidae	White perch	*Morone americana*
	White bass	*Morone chrysops*
Percidae	Yellow perch	*Perca flavescens*
	Sauger	*Sander canadensis*
	Walleye	*Sander vitreus*
Salmonidae	Lake whitefish	*Coregonus clupeaformis*
	Pink salmon	*Oncorhynchus gorbuscha*
	Coho salmon	*Oncorhynchus kisutch*
	Chinook salmon	*Oncorhynchus tshawytscha*
	Rainbow trout	*Oncorhynchus mykiss*
	Atlantic salmon	*Salmo salar*
	Brown trout	*Salmo trutta*
	Lake trout	*Salvelinus namaycush*
Sciaenidae	Freshwater drum	*Aplodinotus grunniens*
Alternative Fish Species		
Family Name	**Common Name**	**Scientific Name**
Catostomidae	Quillback	*Carpiodes cyprinus*
	Longnose sucker	*Catostomus catostomus*
	White sucker	*Catostomus commersonii*
	Northern hog sucker	*Hypentelium nigricans*
	Bigmouth buffalo	*Ictiobus cyprinellus*
	Black buffalo	*Ictiobus niger*
Centrarchidae	Green sunfish	*Lepomis cyanellus*
	Pumpkinseed	*Lepomis gibbosus*
	Warmouth	*Lepomis gulosus*
	Bluegill	*Lepomis macrochirus*
	Longear sunfish	*Lepomis megalotis*
Ictaluridae	Black bullhead	*Ameiurus melas*
	Yellow bullhead	*Ameiurus natalis*
	Brown bullhead	*Ameiurus nebulosus*
Salmonidae	Cisco	*Coregonus artedi*
	Bloater	*Coregonus hoyi*
	Round whitefish	*Prosopium cylindraceum*
	Brook trout	*Salvelinus fontinalis*

Table 7-7. Sampling Procedures for Human Health Fish Tissue Composite Samples

1. Put on clean nitrile gloves before handling the fish. Do not handle any food, drink, sunscreen, or insect repellant until after the composite sample has been collected, measured, and wrapped.
2. Rinse potential target species/individuals in ambient water to remove any foreign material from the external surface and placed in clean holding containers (e.g., livewells, buckets).
3. The composite should consist of 5 fish of adequate size to provide a total of 500 grams of fillet tissue for human health samples. Select fish for each composite based on size similarity and priority target species or alternative fish species in Table 7-5.

   *Accurate taxonomic identification is essential in assuring and defining the organisms that have been submitted for analysis. Individuals from different species should **not** be used in a single sample.*
4. Measure each individual fish to determine total body length. Measure total length of each specimen in millimeters, from the anterior-most part of the fish to the tip of the longest caudal fin ray (when the lobes of the caudal fin are depressed dorsoventrally).
5. On the Human Health Fish Collection Form, side 2 (Figure 7-4), record the sample identification number and response to length and species question in the header of the form. Below the header, record species retained, specimen length (total length in mm), and any relevant comments. Extra rows are provided on the form in the event that additional specimens are collected to meet the 500 gram fillet tissue requirement (refer to Frequently Asked Questions for further clarification). Make sure the sample ID and specimen numbers recorded on the form match those on the sample labels.
6. Remove each fish retained for analysis from the clean holding container(s) (e.g., livewell) using clean nitrile gloves. Dispatch each fish using a clean wooden bat (or equivalent wooden device).
7. Wrap each fish in extra heavy-duty aluminum foil, with the dull side in (foil provided by EPA as solvent-rinsed, oven-baked sheets).
8. Prepare a Sample Identification Label for each sample, ensuring that the label information matches the information recorded on the Human Health Fish Tissue Sample form. **Be sure to include fish species and specimen length on each label.**
9. Cut a length of food grade tubing (provided by EPA) that is long enough to contain each individual fish and to allow extra length on each end to secure with cable ties. Place each foil-wrapped specimen into the appropriate length of tubing. Seal each end of the tubing with a plastic cable tie, and attach the appropriate Sample Identification Label to the plastic tubing using clear tape (wrapping completely around the wrapped fish so that the clear tape wraps over itself).
10. Double-bag the entire set of specimens in the composite, that is, place all the fish in the composite from the site inside a large plastic bag.
11. Prepare a Sample Identification Label for the outer bag, ensuring that the label information matches the information recorded on the Fish Collection Form. **Be sure to include fish species and lengths on the label.** Affix the sample label to a Tyvek tag and cover with clear plastic tape. Thread a cable tie through the grommet in the Tyvek tag and seal the outer bag with the cable tie
12. As each sample is packaged, place it immediately on dry ice for shipment. If samples will be carried back to a laboratory or other facility to be frozen before shipment, wet ice can be used to transport wrapped and bagged fish samples in the coolers to a laboratory or other interim facility.
13. If possible, keep all specimens designated for a particular composite in the same shipping container (ice chest) for transport.
14. Samples may be stored on dry ice for a maximum of 24 hours. You have the option, depending on site logistics, of:
    - shipping the samples packed on sufficient quantities of dry ice (50 pounds minimum, layered to ensure direct contact between fish and dry ice) to keep samples frozen for up to 48 hours, via priority overnight delivery service (e.g., Federal Express), so that they arrive at the sample preparation laboratory within less than 24 hours from the time of sample collection, or
    - freezing the samples within 24 hours of collection at ≤-20°C, and storing the frozen samples until shipment within 2 weeks of sample collection. Frozen samples will subsequently be packed on at least 50 pounds of layered dry ice and shipped to the sample preparation laboratory via priority overnight delivery service.



NCCA 2010 HUMAN HEALTH FISH COLLECTION (Front) Reviewed by (initial): JPS

SITE ID: NCCAGL10 MI0000 DATE: 06/01/2010

Collection Method

○ TRAWL ○ HOOK & LINE ○ SEINE
● GILL NET
○ OTHER EXPLAIN:

Trawl/Seine Info: HELMSMAN: LINE OUT (m):

Trawl/Seine Start:
LATITUDE: LONGITUDE: HEADING DEGREES MAGNETIC: START TIME:
Decimal Degrees
FLAG:

Trawl/Seine End:
LATITUDE: LONGITUDE: END TIME:
Decimal Degrees
FLAG:

If Seine: LENGTH: (m) FLAG:

If Gill Net: LENGTH: 50 (m) START TIME: 08 00 END TIME: 13 30 FLAG:

Flag Codes: K = no measurement made; U = suspect measurement; F1, F2, etc. = flags assigned by each field crew. Explain all flags in comments section.

Flag	Comments

DRAFT NOT FOR USE

03/31/2010 NCCA Human Health Fish Collection (Front) 5721053452

Figure 7-3. Example Human Health Fish Tissue Data Form (Front)

NCCA 2010 HUMAN HEALTH FISH COLLECTION (Back) Reviewed by (initial): JPS

SITE ID: NCCAGL10 MI0000 DATE: 06/01/2010 PAGE: 1 OF 1

FISH TISSUE SAMPLE ○ NO SAMPLE COLLECTED

SAMPLE ID 999110 FISH ALL WITHIN 75% OF LARGEST SPECIMEN ● FISH ARE ALL THE SAME SPECIES ●

	Genus Species	Total Length (mm)	Frozen	Comments
.1	SANDER VITREUS	403	●	
.2		350	●	
.3		441	●	
.4		425	●	
.5		464	●	
			○	
			○	
			○	
			○	
			○	

Human health fish composites should consist of 5 similarly-sized adult fish of the same species that will collectively yield about 500 g of fillet tissue (refer to Frequently Asked Questions for possible exceptions).

7461152602

03/31/2010 NCCA Human Health Fish Collection (Back)

Figure 7-4. Example Human Health Fish Tissue Data Form (Back)

8.0 FINAL SITE ACTIVITIES

Prior to leaving the site, make a general visual assessment of the site and its adjacent shoreline. The objective of the site assessment is to record observations of the shoreline and site characteristics that are useful for future data interpretation, ecological value assessment, development of associations, and verification of stressor data. Your observations and impressions are extremely valuable.

You will filter and process the fecal indicator and chlorophyll-*a* samples, and collect the dissolved nutrients sample from the chlorophyll-*a* filtrate. You will also conduct a final check of the data forms, labels and samples. The purpose of the second check of data forms, labels and samples is to assure completeness of all sampling activities. Finally, clean and pack all equipment and supplies, and clean the launch site and staging areas. After you leave the site, report the sampling event to the Information Management Coordinator, and ship or store the samples. Activities described in this section are summarized in Figure 8-1.

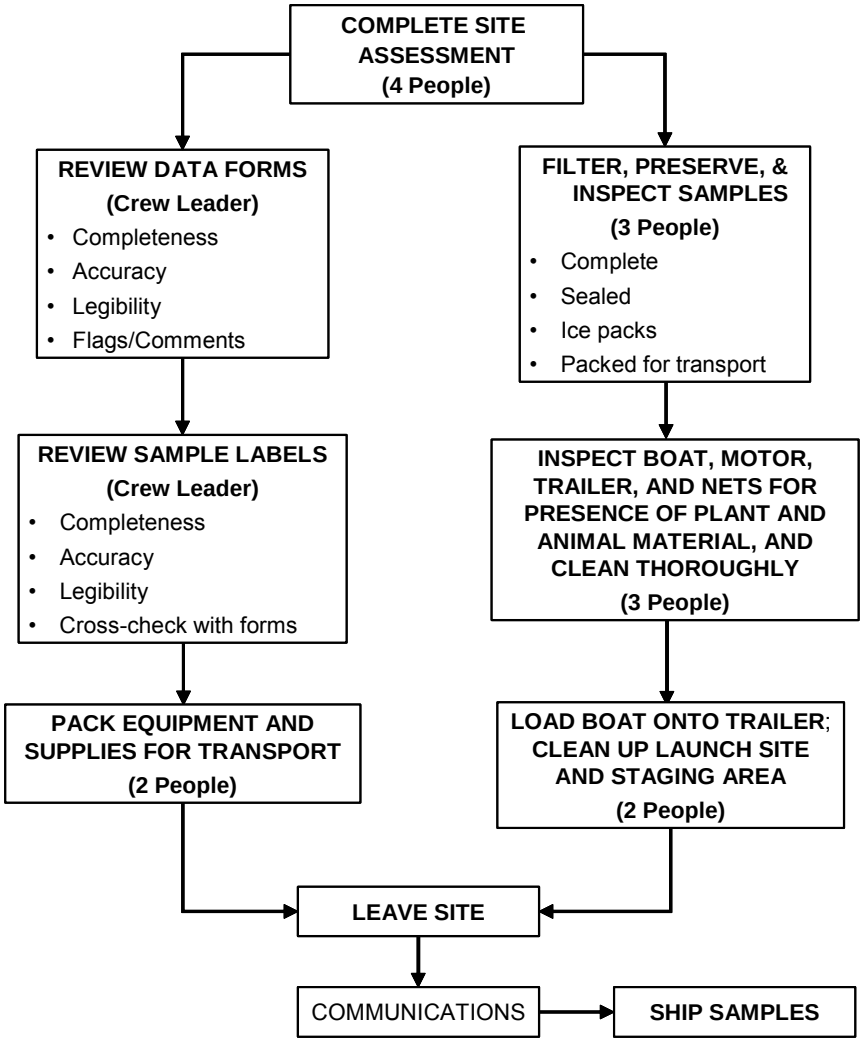


Figure 8-1. Final Site Activities Summary.

8.1 General Site Assessment

Complete the Site Assessment Form (Figure 8-2) after sampling, recording all observations from the site that were noted during the course of the visit. This Site Assessment Form is designed as a template for recording pertinent field observations. It is by no means comprehensive, and any additional observations should be recorded in the General Assessment section.

8.1.1 Shoreline Activities and Disturbances

Record shoreline activities and disturbances on a rating of low, moderate or high. For this portion of the site assessment, consider the shoreline adjacent to the sampling site that is visible from the X-site. Consider only the shoreline which is in the same estuary, waterbody and/or embayment as the X-site. Using your best judgment, direct the assessment to the shoreline that would be considered ecologically significant to the sampling site. If the shore cannot be seen from the X-site (due to weather conditions or distance) note in the comments section the reason that the shoreline assessment was not possible.

8.1.2 Site Characteristics

Record observations regarding the general characteristics of the site on the Site Assessment Form. When assessing these characteristics, look at a 200 m radius around the X-site. Rank the site between “pristine” and “highly disturbed”, and between “appealing” and “unappealing” on a scale of 1 to 5. As with other aspects of the general visual assessment, all crew members should provide input into the final ranking. These observations will be understandably subjective, but provide valuable information on crew impressions of the overall character of the site that can be used by NCCA analysts to help explain data. Document any signs of pipe outflows or shoreline enhancement or retention. Document the weather conditions on the day of sampling, and any extreme weather conditions just prior to sampling.

8.1.3 General Assessment

Record any additional information and observations in this narrative section. Information to include could be observations on biotic integrity, presence of SAV, presence and abundance of endangered and/or exotic species, local anecdotal information, or any other pertinent information about the site or its adjacent areas. Record any observations that may be useful for future data interpretation.

NCCA 2010 SITE ASSESSMENT (Front)

Reviewed by (initial): JPS

SITE ID: NCCA10 LA0000 DATE: 06/01/2010

SHORELINE ACTIVITIES AND DISTURBANCES OBSERVED (Intensity: Blank=Not observed, L=Low, M=Moderate, H=Heavy)

Residential	Recreational	Agricultural	Industrial	Management
L M H Residences	L M H Hiking Trails	L M H Cropland	L M H Industrial Plants	L M H Chemical Treatment
L M H Maintained Lawns	(L) M H Parks, Campgrounds	L M H Pasture	L M H Mines/Quarries	(L) M H Angling Pressure
L M H Construction	L M H Primitive Parks, Camping	L M H Livestock Use	L M H Oil/Gas Wells	L M H Dredging
L M H Pipes, Drains	L M H Trash/Litter	L M H Orchards	L M H Power Plants	L M H Channelization
L M H Dumping	L M H Surface Films	L M H Poultry	L M H Logging	L M H Water Level Fluctuations
L M H Roads	L M H Dunes	L M H Irrigation Equip.	L M H Evidence of Fire	L M H Shoreline Hardening
L M H Bridge/Culverts	(L) M H Beach	L M H Water Withdrawal	L M H Odors	L M H Dredge Material
L M H Sewage Treatment	L M H Forested		L M H Commercial	

SITE CHARACTERISTICS (200 m radius)

Waterbody Character	Pristine	O 5	O 4	● 3	O 2	O 1 Highly Disturbed
	Appealing	O 5	● 4	O 3	O 2	O 1 Unappealing

Dominant Land Use

Dominant Land Use Shoreline: O Forest O Agriculture O Range O Urban O Suburban/Town

If Forest, Dominant Age Class: O 0 - 25 yrs. O 25 - 75 yrs. O > 75 yrs.

WEATHER PARTLY CLOUDY, WAVES LESS THAN 1 FOOT, WIND OUT OF THE SOUTH. VISIBILITY AROUND 10 MILES

GENERAL ASSESSMENT (Biotic integrity, Vegetation diversity, Local anecdotal information)

CANNOT MAKE ACCURATE SHORELINE OBSERVATIONS, CLOSEST POINT OF LAND IS 2.6 NAUTICAL MILES TO THE EAST. GULLS AND TERNS OBSERVED AT THE INDEX SITE. BOTTOM IS SOFT MUCK AT THIS SITE. IN ADDITION TO THE PINFISH COLLECTED FOR THE ECO FISH SAMPLE, WE COLLECTED 2 HARDHEAD CATFISH, 1 ATLANTIC CROAKER, AND 1 SHRIMP. OBSERVED 1 RECREATIONAL FISHING BOAT WHILE TRAVELING TO THE INDEX SITE.

7216195652

03/31/2010 NCCA Site Assessment

Figure 8-2. Example Site Assessment Form.

8.2 Processing The Fecal Indicator And Chlorophyll-*a* Samples

8.2.1 Equipment and Supplies (Fecal Indicator)

Table 8-1 provides the equipment and supplies needed for field crews to filter the fecal indicator sample. The filtering apparatus for this indicator MUST be sterile.

Table 8-1. Equipment and Supplies List for Fecal Indicator Sample

For processing samples	▪ Nitrile gloves ▪ Sterile screw-cap 50 mL tube ▪ Sterile filter holder, Nalgene 145/147 ▪ Vacuum pump (electric or hand) ▪ Sterile saline ▪ Whatman 47 mm polycarbonate 0.4 µm sterile filters ▪ Sterile disposable forceps ▪ 4 sterile microcentrifuge tubes containing sterile glass beads (chilled on dry ice during pre-sampling activities) ▪ Sterile 60 X 15 mm Petri dish ▪ Dry ice ▪ Cooler ▪ Field Operations Manual or laminated Quick Reference Guide
For recording measurements	▪ Sample Collection Form ▪ Soft (#2) lead pencils for recording data on field forms ▪ Fine-tipped indelible markers for filling out sample labels ▪ Fecal Indicator sample labels (4 vial labels and 1 bag label) ▪ Clear tape strips for covering labels

8.2.2 Procedures for Processing the Fecal Indicator Sample

The fecal indicator sample **must** be filtered **before** the chlorophyll-*a* samples, since the filtering apparatus needs to be sterile for this sample. The procedures for processing the fecal indicator sample are presented in Table 8-2. The sample must be filtered and frozen within 6 hours of collection.

At revisit sites, a filter blank is prepared **prior** to filtering the Enterococci sample. Filter blanks will be prepared at both Visit 1 and Visit 2. Procedures for preparing the filter blank are presented in Table 8-3.

Table 8-2. Processing Procedure—Fecal Indicator Sample

1. Put on nitrile gloves.
2. Set up sample filtration apparatus on flat surface and attach vacuum pump. Set out 50 mL sterile centrifuge tube, sterile 60 mm Petri dish, 1 bottle of chilled sterile saline, Whatman 47 mm polycarbonate sterile filter box, and 2 sterile forceps.
3. Aseptically transfer 4 polycarbonate filters from filter box to base of opened Petri dish. Close the filter box and set aside.
4. Remove cellulose nitrate (CN) filter (the filter with grid design on it) from funnel and discard. Be sure to leave the support pad in the filter funnel.
5. Load filtration funnel with sterile polycarbonate filter on support pad (shiny side up). Be sure not to use the blue divider paper as the filter.
6. Shake sample bottle(s) 25 times to mix well.
7. Measure 25 mL of the mixed water sample in the sterile graduated centrifuge tube and pour into filter funnel.
8. Replace cover on filter funnel and pump to generate a vacuum (do not generate more than 7 inches of Hg of vacuum ~15 psi). Keep pumping until all liquid is in filtrate collection flask.
9. If the first 25 mL volume passes readily through the filter, add another 25 mL and continue filtration. If the filter clogs before completely filtering the first or second 25 mL volume, discard the filter and repeat the filtration using a lesser volume.
10. Pour approx. 10 mL of the chilled sterile saline into the graduated centrifuge tube used for the sample. Cap the tube and shake 5 times. Remove the cap and pour the rinse into filter funnel to rinse filter.
11. Filter the rinsate and repeat with another 10 mL of sterile saline.
12. Remove filter funnel from base without disturbing filter. Using sterile disposable forceps remove the filter (touching only the filter edges) and fold it in half, in quarters, in eighths, and then in sixteenths (filter will be folded 4 times).
13. Insert filter into chilled filter extraction tube (with beads) open end first (pointy end up). Replace and tighten the screw cap, insert tube(s) into bubble envelope on dry ice for preservation during transport and shipping.
14. Record the volume of water sample filtered through the filter (minimum is 25 mL, target is 50 mL) and the volume of saline rinse each filter was rinsed with on the Sample Collection Form, Side 1. Record the filtration start time (beginning of first filter) and finish time (end of fourth filter) for the sample.
15. Repeat steps 6 to 15 for the remaining three filters. It is important that the same sample volume be filtered through each of the 4 filters.

Table 8-3. Processing Procedure—Fecal Indicator Filter Blank

Enterococci filter blanks will be prepared at all revisit sites during both visit 1 and visit 2. **Prepare the filter blanks before filtering the coastal sample**.

1. Set up sample filtration apparatus using same procedure as used for the sample above. Chill Filter Extraction tubes with beads on dry ice.
2. Aseptically transfer 4 polycarbonate filters from filter box to base of opened Petri dish. Close filter box and set aside.
3. Remove cellulose nitrate (CN) filter (the filter with grid design on it) from funnel and discard. Be sure to leave the support pad in the filter funnel.
4. Load filtration funnel with sterile polycarbonate filter on support pad (shiny side up).
5. Measure 20 mL of the chilled sterile saline in the sterile graduated centrifuge tube and pour into filter funnel.
6. Replace cover on filter funnel and pump to generate a vacuum (do not generate more than 7 inches of Hg of pressure). Keep pumping until all liquid is in filtrate collection flask.
7. Remove filter funnel from base without disturbing filter. Using sterile disposable forceps remove the filter (touching only the filter edges) and fold it in half, in quarters, in eighths, and then in sixteenths (filter will be folded 4 times).
8. Insert filter into chilled filter extraction tube (with beads) open end first (pointy end up). Replace and tighten the screw cap, insert tube(s) into bubble envelope on dry ice for preservation during transport and shipping.
9. Record the filter blank information on the Sample Collection Form.
10. Label the samples as "blank" on the label and field form, and package and submit these samples to the lab with the standard samples.
11. Repeat steps 4 to 9 for the remaining three 20 mL volumes of sterile saline to be filtered.

8.2.3 Equipment and Supplies (Chlorophyll-*a* and Dissolved Nutrients Sample)

Table 8-4 provides the equipment and supplies needed to process the chlorophyll-*a* sample.

Table 8-4. Equipment and Supplies List for Chlorophyll-*a* and Dissolved Nutrients Processing

For filtering chlorophyll-*a* sample	▪ Whatman GF/F 47mm 0.7 micron filter pads ▪ Filtration apparatus with graduated filter holder ▪ Vacuum pump (electric or hand)	▪ DI water ▪ Nitrile gloves ▪ Forceps ▪ Graduated cylinders
For recording measurements	▪ Sample Collection Form ▪ Sample labels ▪ #2 pencils	▪ Fine-tipped indelible markers ▪ Clear tape strips
For sample collection and preservation	▪ 50 mL screw-top centrifuge tube ▪ Aluminum foil square ▪ Plastic (electrical) tape	▪ Whirl-pak ▪ Cooler with dry ice ▪ 250 mL Nalgene bottle

8.2.4 Procedures for Processing the Chlorophyll-*a* and Dissolved Nutrients Sample

The procedures for processing chlorophyll-*a* and dissolved nutrients samples are presented in Table 8-5. Whenever possible, sample processing should be done in subdued light, out of direct sunlight.

Table 8-5. Processing Procedure—Chlorophyll-*a* and Dissolved Nutrients Sample

1. Put on nitrile gloves.
2. Discard any filtrate collected in the filter flask during the filtration of the Enterococci sample and rinse the flask thoroughly (three times) with DI water.
3. Rinse the filter funnel three times with DI water prior to filtration. Rinse graduated cylinders with DI water.
4. Use clean forceps to place a Whatman GF/F 47 mm 0.7 micron filter in the graduated filter holder apparatus (the same apparatus used previously for filtering the Enterococci sample) with the gridded side of the filter facing down.
5. Remove chlorophyll-*a* collection bottle from cooler and shake to mix sample. Pour 250 mL of water into the filter holder, replace the cap, and use the vacuum pump to draw a small portion of the sample through the filter.
6. Use the first 10-20 mL of filtrate to rinse the inside of the filter flask and discard the rinsate. Replace the filter flask and continue filtering. Repeat with an additional two rinses of filtered site water.
7. If 250 mL of site water will not pass through the filter, change the filter, rinse the apparatus with DI water, and repeat the procedures using 100 mL of site water. **Do not exceed 7 inches of Hg of vacuum ~15 psi or a filtration duration of more than 5 minutes for a single sample volume, to avoid cell damage or loss of contents during filtering.**
8. Observe the filter for readily visible color. If there is visible color, proceed; if not, filter additional aliquots until color is visible on the filter or until a maximum of 2,000 mL have been filtered.
9. After collecting at least 250 mL of filtrate in the filter flask, remove the bottom portion of the apparatus and pour 250 mL of the filtrate into the 250 mL Nalgene bottle. This filtrate is used for dissolved nutrient analyses. Complete the label, including salinity (as taken during the *in situ* measurements) and affix to the 250 mL Nalgene bottle. Cover with clear plastic tape
10. Record the dissolved nutrients sample information on the Sample Collection Form. Place the sample on wet ice.
11. After achieving a readily visible stain on the filter, remove the filter from the holder with clean forceps. Avoid touching the colored portion of the filter. Fold the filter in half, with the colored side folded in on itself. Place into a 50 mL screw-top centrifuge tube.
12. Complete the label, including volume filtered and affix to the 50 mL screw-top centrifuge tube. Cover with clear plastic tape. Record the actual sample volume filtered on the Sample Collection Form.
13. Seal the cap of the centrifuge tube with plastic tape.
14. Wrap the 50 mL tube in a foil square and place in a whirl-pak.
15. Place the whirl-pak containing the filter on dry ice.

8.3 Data Forms And Sample Inspection

After the Site Assessment Form is completed, the Field Team Leader reviews all of the data forms and sample labels for accuracy, completeness, and legibility. The other team members inspect all sample containers and package them in preparation for transport, storage, or shipment. Refer to Appendix D for details on preparing samples for shipping.

Ensure that all required data forms for the site have been completed. Confirm that the SITE-ID, the visit number, and date of visit are correct on all forms. On each form, verify that all information has been recorded accurately, the recorded information is legible, and any flags are explained in the comments section. Ensure that written comments are legible, with no "shorthand" or abbreviations. Make sure there are no stray markings in on the forms. Make sure the header information is completed on all pages of each form. After reviewing each form, initial the upper right corner of each page of the form.

Ensure that all samples are labeled, all labels are completely filled in, and each label is covered with clear plastic tape. Compare sample label information with the information recorded on the corresponding field data forms (e.g., the Sample Collection Form) to ensure accuracy. Make sure that all sample containers are properly sealed.

8.4 Launch Site Cleanup

Load the boat on the trailer and inspect the boat, motor, and trailer for evidence of weeds and other macrophytes. Clean the boat, motor, and trailer as completely as possible before leaving the launch site. Follow any state or other requirements associated with nuisance species, pathogens and/or viruses. Inspect all nets for pieces of macrophytes or other organisms and remove as much as possible before packing the nets for transport. Pack all equipment and supplies in the vehicle and trailer for transport. Keep equipment and supplies organized so they can be inventoried using the equipment and supply checklists presented in Appendix A. Lastly, be sure to clean up all waste material at the launch site and dispose of or transport it out of the site if a trash can is not available.

9.0 FIELD QUALITY CONTROL

Standardized training and data forms provide the foundation to help ensure that data quality standards for field sampling are met. These Standard Operating Procedures for field sampling and data collection are the primary guidelines for all cooperators and field teams. In addition, repeat sampling, and field evaluation and assistance visits will address specific aspects of the data quality standards for the National Coastal Condition Assessment.

9.1 Repeat Sampling

Repeat sampling will provide data to make variance estimates (for measurement variation and index period variation) that can be used to evaluate the NCCA design for its potential to estimate status and detect trends in the target population of sites. The sites identified for repeat visits are outlined in the site list provided to each state.

A total of 10% of the target sites visited will be revisited during the same field season by the same field team that initially sampled the site. Repeated samples and measurements are taken from the same site as the first visit. Each state has a different number of repeat sites; the number is dependent on the number of base sites the state has and is provided to each state in the state's site draw. If a site selected for repeat sampling is dropped, then the alternate assigned to replace it should be revisited. The primary purpose of this "revisit" set of sites is to collect temporal replicate samples to provide variance estimates for both measurement variation and index period variation. The revisit will include the full set of indicators and associated parameters (except fish tissue). We will not be collecting replicate data on fish tissue. Fish tissue will only be collected on the first visit. The time period between the initial and repeat visit to a site should be as long as possible, but not less than 2 weeks.

In addition to the normal samples, filter blanks will be collected for Enterococci on both of the two visits. The teams will filter a small amount (20 mL) of sterile saline through 4 filters, label them and write "blank" on the label and field form, and package and submit these samples to the lab. The filter blanks should be run **before** the sample is filtered. A detailed description of the filter blanks is found in table 8-3.

9.2 Field Evaluation And Assistance Visits

A rigorous program of field and laboratory evaluation and assistance visits has been developed to support the National Aquatic Resource Surveys Program. These evaluation and assistance visits are explained in detail in the Quality Assurance Project Plan (QAPP) for the NCCA. The following sections will focus only on the field evaluation and assistance visits.

These visits provide a QA/QC check for the uniform evaluation of the data collection methods, and an opportunity to conduct procedural reviews as required to minimize data loss due to improper technique or interpretation of field procedures and guidance. Through uniform training of field teams and review cycles conducted early in the data collection process, sampling variability associated with specific implementation or interpretation of the protocols will be significantly reduced. The field evaluations will be based on the Field Evaluation Plan and Checklists. This evaluation will be conducted for each unique team collecting and contributing data under this program (EPA will make a concerted effort to evaluate every team, but will rely

on the data review and validation process to identify unacceptable data that will not be included in the final database).

9.2.1 Specifications for QC Assurance

Field evaluation and assistance personnel are trained in the specific data collection methods detailed in this Field Operations Manual. A plan and checklist for field evaluation and assistance visits have been developed to detail the methods and procedures. The plan and checklist are included in the QAPP. Table 9-1 summarizes the plan, the checklist, and corrective action procedures.

Table 9-1. General Information Noted During Field Evaluation

Field Evaluation Plan	▪ Regional Coordinators will arrange the field evaluation visit with each Field Team, ideally within the first two weeks of sampling. ▪ The Evaluator will observe the performance of a team through one complete set of sampling activities. ▪ If the Team misses or incorrectly performs a procedure, the Evaluator will note it on the checklist and immediately point it out so the mistake can be corrected on the spot. ▪ The Evaluator will review the results of the evaluation with the Field Team before leaving the site, noting positive practices and problems.
Field Evaluation Checklist	▪ The Evaluator observes all pre-sampling activities and verifies that equipment is properly calibrated and in good working order, and NCCA protocols are followed. ▪ The Evaluator checks the sample containers to verify that they are the correct type and size, and checks the labels to be sure they are correctly and completely filled out. ▪ The Evaluator confirms that the Field Team has followed NCCA protocols for locating the site. ▪ The Evaluator observes the complete set of sampling activities, confirming that all protocols are followed. ▪ The Evaluator will record responses or concerns, if any, on the Field Evaluation and Assistance Check List.
Corrective Action Procedures	▪ If the Evaluator's findings indicate that the Field Team is not performing the procedures correctly, safely, or thoroughly, the Evaluator must continue working with this Field Team until certain of the Team's ability to conduct the sampling properly so that data quality is not adversely affected. ▪ If the Evaluator finds major deficiencies in the Field Team operations, the Evaluator must contact a NCCA QA official immediately (e.g., within 24-48 hours) so that additional correction actions can be taken.

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. If the Field Team misses or incorrectly performs a procedure, the Evaluator will note this on the checklist and immediately point this out so the mistake can be corrected on the spot. The role of the Evaluator is to provide additional training and guidance so that the procedures are being performed consistent with the Field Operations Manual, all data are recorded correctly, and paperwork is properly completed at the site.

9.2.2 Reporting

When the sampling operation has been completed, the Evaluator will review the results of the evaluation with the Field Team before leaving the site (if practicable), noting positive practices and problems (i.e., weaknesses [might affect data quality] or deficiencies [would adversely affect data quality]). The Evaluator will ensure that the Team understands the findings and will be able to perform the procedures properly in the future. The Evaluator will record responses or concerns, if any, on the Field Evaluation and Assistance Check List. After the Evaluator completes the Field Evaluation and Assistance Check List, including a brief summary of findings, all Field Team members must read and sign off on the evaluation.

If the Evaluator's findings indicate that the Field Team is not performing the procedures correctly, safely, or thoroughly, the Evaluator must continue working with this Field Team until certain of the Team's ability to conduct the sampling properly so that data quality is not adversely affected. If the Evaluator finds major deficiencies in the Field Team operations (e.g., major misinterpretation of protocols, equipment or performance problems) the Evaluator must contact the following QA official:

- Joe Hall, EPA National Coastal Condition Assessment Project QA Officer

The QA official will contact the Project Manager to determine the appropriate course of action. Data records from sampling sites previously visited by this Field Team will be checked to determine whether any sampling sites must be redone.

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Web Pages:

US EPA Aquatic Monitoring Research: http://www.epa.gov/nheerl/arm
NHD Plus: http://www.horizon-systems.com/nhdplus

APPENDIX A

List of Equipment and Supplies

EQUIPMENT & SUPPLY LISTS

General Equipment

- Field Operations Manual and/or laminated Quick Reference Guide
- Covered clipboards
- Field forms and sample labels
- Clear tape strips for covering labels
- Pencils (#2)
- Fine-tipped indelible markers
- Digital camera with extra memory card & battery
- Maps and access instructions
- Sampling permits and/or permission letters
- GPS unit with manual and reference card
- Batteries
- 1% - 10% Bleach
- Calibration cups and standards for multi-probe unit
- Spare parts for multi-probe unit
- Electrical and duct tape
- Scissors
- Plastic storage tub
- Cell phone, 2-way radios, and/or walkie-talkies
- Centimeter ruler

Boat Equipment List

- Motor
- Gas Can
- PFDs (1/person)
- Type IV PFD (Throwable Life Saving device)
- Bow/Stern lights
- Anchor with 75 m line or sufficient to anchor in 50 m depth
- Float to attach to anchor
- Sonar Unit
- Pingers
- First Aid Kit
- Extra Boat Plug
- Spare Prop Shear Pin
- Emergency Tool kit
- Hand Bilge pump
- Fire Extinguisher
- Boat horn
- Spare prop

Sample/Data Collection

- Multi-parameter water quality meter with pH, DO, temperature, and conductivity/salinity probes (e.g., Hydrolab, YSI, etc.)
- 20 cm diameter Secchi disk and calibrated sounding line, marked in 0.5 m intervals
- Nitrile gloves
- Water sampling bottle (e.g., Niskin) or sampling pump system
- PAR meter with appropriate deck unit and cables
- Thermometer
- 0.04 m2 Young-modified Van Veen grab sampler or Standard or Petite Ponar sampler with plastic tub, drop line, and spare pinch pin.
- Weights and pads for grab
- 0.5 mm stainless steel sieve; 1.0 mm for CA, OR, and WA
- Sieve box
- Plastic tub or bucket for benthic sediment grab
- High-quality stainless mixing pot with lid
- Large and small stainless steel spoons and spatulas for mixing and dispensing sediment composite
- Wide-mouthed funnel
- Fine-tipped forceps
- Large buckets
- Electrical tape
- Scrub brush
- Squirt bottle
- Measuring board (millimeter scale)
- Pre-sterilized, 250 mL sample bottle
- Sodium thiosulfate tablet
- De-ionized water in squirt bottle
- Active or passive fish sampling device (e.g., trawl, seine, hook & line, etc.)

Sample Processing/Preservation

- Coolers
- Wet ice
- Dry ice
- 100% buffered formalin with Stain
- Sterile filtration unit (Nalgene 145/147), including filter funnel, cap, filter holder, and receiving chamber
- Whatman 47 mm polycarbonate 0.4 micron filters
- Whatman 47 mm 0.7 micron GF/F glass fiber filters
- Sterile disposable forceps
- Sterile saline
- Wooden bat
- Alconox
- Aluminum foil squares (3” x 6”)
- DI water
- Small stainless steel spatula, spoon, or scoop to transfer sample
- Knife or scissors
- Plastic cable ties
- Sterile filter holder
- Hand or electric vacuum pump
- Sterile centrifuge tube
- 60 x 15 disposable Petri dishes sterile microcentrifuge tubes containing sterile glass beads

Sample Storage

- 250 mL amber Nalgene bottle (water chemistry)
- 2 L amber Nalgene bottle (chlorophyll-*a*)
- 250 mL sterile sample bottle (Enterococci)
- 1 L Nalgene bottles (benthic samples)
- 250 mL Nalgene bottle (dissolved nutrients)
- 500 mL glass jar (sediment organics/metals)
- 125 mL Nalgene jar (sediment grain size)
- 60 mL Nalgene jar (sediment TOC)
- 1 gallon screw-top bucket (sediment toxicity)
- 50 mL screw-top centrifuge tube
- Coolers
- Whirl-Paks
- 2 gallon self-sealing bags (eco fish tissue)
- Solvent rinsed aluminum foil, poly tubing and zip ties (human health fish tissue)
- Large plastic composite bags

Packaging/Shipping

- Coolers
- Cooler liners (30 gal garbage bags)
- Dry ice (~50 lbs per site)
- Wet ice (~50 lbs per site; additional for shipping)
- Self-sealing bags
- Packing/strapping tape
- FedEx airbills
- Class 9 Dangerous Goods label (for dry ice shipments)

Sample Collection/Preservation of Additional Great Lakes Indicators

- 1 L amber Nalgene bottle (phytoplankton)
- Underwater camera system with DVR and GPS
- Lugol's solution
- 10 mL pipet and pipet bulb

A **site kit** will be provided to the field crews for each sampling site. Site kits will be shipped out based on the schedule that each field crew provides prior to the start of the sampling season. **Field crew leaders MUST provide a schedule in order to receive the site kits.** If your schedule changes, please report the change as soon as possible to the Site Kit Coordinator and/or Field Logistics Coordinator. Prior to sampling, inspect each site kit to ensure all supplies are included.

Supplies provided in each Site Kit:

- Field Data Forms
- Sample Labels
- National Coastal Condition Assessment Fact Sheets
- 1 250 mL amber Nalgene bottle (water chemistry)
- 2 1 L Nalgene bottles (benthic samples)
- 1 500 mL glass jar (sediment organics/metals)
- 1 60 mL Nalgene jar (sediment TOC)
- 1 125 mL Nalgene jar (sediment grain size)
- 1 1 gallon screw-top bucket (sediment toxicity)
- 1 250 mL Nalgene bottle (dissolved nutrients)
- 1 250 mL fecal indicator collection bottle
- 1 Zip tie
- 2 50 mL screw-top centrifuge tube (one for measuring enterococci sample for filtering, one for holding the chlorophyll-*a* filter)
- 4 sterile microcentrifuge tubes containing sterile glass beads
- Bubble envelope
- 2 Sterile disposable forceps
- Sterile filter holder, Nalgene 145/147
- Sterile saline
- Large plastic bag (cooler liner)
- FedEx airbills for all labs
- Dry ice box/label will be included in approximately every 4th site kit
- Whirl-Paks

Supplies provided in each Eco Fish Tissue Sampling Kit:

- 2 gallon self sealing bags
- Sandwich size self sealing bags
- Large plastic (composite) bags
- Plastic cable ties
- Tyvek tags with grommets
- FedEx airbill for fish tissue lab

Supplies provided in each Human Health Fish Tissue Sampling Kit:

- Aluminum foil (solvent-rinsed and baked)
- Heavy-duty food grade polyethylene tubing
- Large plastic (composite) bags
- Plastic cable ties
- Tyvek tags with grommets
- FedEx airbill for fish tissue lab

Supplies provided in each Base Kit:

- 1 2 L amber Nalgene bottle (chlorophyll)
- Nitrile gloves
- Bottle of 50 sodium thiosulfate tablets
- Aluminum foil 3x6”
- 15” stainless steel spoon
- 0.5 mm stainless steel sieve bucket; 0.1 mm for CA, OR, WA sampling
- Weighted Secchi disk
- 2 1 Liter Nalgene wash bottles
- 1 3 gallon Rubbermaid Roughneck tote
- Graduated cylinder 250 mL
- Whatman 47 mm polycarbonate 0.4 µ filters
- Whatman 47 mm glass fiber GF/F 0.7 µ filters
- Hand pump
- Silicone stopper with filter holder adapter
- Spare filter holder adapters
- Side-arm filter flask
- Disposable petri dishes 60x15
- Wide-mouthed funnel
- Fine-tipped forceps
- Centrifuge tube stand
- 1 liter of QC check solution (re-order as expiration date approaches)
- Tape dispenser
- Tape strips
- 24 ct of 1 Liter Nalgene bottles
- Whirl-Paks
- Heavy-duty food grade polyethylene tubing and zip ties (for large Eco fish)
- 12 Spare 2 gallon self sealing bags
- Spare sterile saline
- Spare 50 mL screw-top centrifuge tubes
- Spare sterile microcentrifuge tubes
- Spare Sterile filter holder
- 1 qt Self Sealing Bags (100 count)

Note: sodium thiosulfate tablets, calibration QC check solution, filters, 1 Liter Nalgene bottles, aluminum foil squares, whirl paks, and disposable nitrile gloves will be provided in the base kit; you may order more throughout the field season if needed.

Additional Base Kit supplies provided to Great Lakes crews:

- Underwater camera system with DVR and GPS
- 1 liter amber Nalgene bottles (approximately 1 per site on sampling schedule)
- 500 mL Lugol’s solution
- 10 mL pipette and pipette bulb

APPENDIX B

Field Forms

Reviewed by (initial): _____

NCCA 2010 SITE VERIFICATION (Front)

SITE NAME: _____ DATE: ___/___/2010 VISIT: ○ 1 ○ 2

SITE ID: NCCA10- STATE OF SITE ____ STATION DEPTH(m): _____ TEAM: _____

DID YOU SAMPLE THIS SITE?

○ YES If YES, check one below

SAMPLEABLE (Choose method used)
- ○ Marine
- ○ Great Lakes

ARRIVAL TIME: ____:____

DEPART TIME: ____:____

○ NO If NO, check one below

NON-SAMPLEABLE-PERMANENT-Replace Site
- ○ Map Error
- ○ Site too shallow for navigation/sampling
- ○ Unsafe

NON-SAMPLEABLE-TEMPORARY-Reschedule
- ○ No Access
- ○ Temporarily Inaccessible-Fire, etc.
- ○ Other (Explain in comments)

VERIFICATION INFORMATION

Site verified by (fill in all that apply): ○ GPS ○ Local Contact ○ Signs ○ Roads ○ Topo. Map
○ Other (Describe Here): ○ Not Verified (Explain in Comments)

LOCATION

Coordinates	Latitude North	Longitude West	# of Satellites	X-SITE WITHIN 37M?:
TARGET Decimal Degrees	____.____	____.____	○ ≤3	○ YES ○ NO
ACTUAL Decimal Degrees	____.____	____.____	○ ≥4	GPS Datum Used (e.g. NAD83,WGS84): ____

HABITAT TYPE: ○ Tidal River ○ Open Water ○ Marsh/Wetland ○ Embayment ○ Inter-Tidal ○ Rivermouth
○ Other, explain: _____

BOTTOM TYPE: ○ Coral Reef ○ Oyster Bed ○ Grass Bed ○ Sand ○ Rocky/Shell ○ Hardpan ○ Mud
○ Other, explain: _____

Debris Present?: ○ YES ○ NO | If Yes, TYPE: ○ Glass ○ Plastic ○ Wood ○ Cans ○ Other, explain: _____

SAV Present?: ○ Yes ○ No ABUNDANCE: _____ (Sparse, dense, etc)

Macroalgae Present?: ○ Yes ○ No ABUNDANCE: _____ (Sparse, dense, etc)

GENERAL COMMENTS:

DIRECTIONS TO SITE:

NCCA 2010 SITE VERIFICATION (Back)

Reviewed by (initial): ____

SITE NAME: _____ DATE: ___/___/2010 VISIT: ○ 1 ○ 2

SITE ID: NCCA10-_____ TEAM: _____

SKETCH MAP - Arrow Indicates North; Mark site L=Launch X=Index F = Fishing Area
NOTE: If an outline map is attached here, use a continuous strip of clear tape across the top edge.
You can also attach a separate sheet with the outline map on it.

PERSONNEL

NAME	Bio/Chem Sampling	Fish	Forms Review
_____	○	○	○
_____	○	○	○
_____	○	○	○
_____	○	○	○
_____	○	○	○

NCCA 2010 FIELD MEASUREMENT (Front)

Reviewed by (initial): _____

SITE ID: NCCA10- _____ DATE: ___/___/2010

CALIBRATION INFORMATION

Instrument manufacturer and model: _____

Instrument ID number: _____ Operator: _____

TEMPERATURE	Thermometer Reading (°C) ___.___	Sensor Reading (°C) ___.___	Flag	Comments
DO	Barometric Pressure (mm Hg) _____	Calibration Value _____.___ O mg/L O %	Displayed Value _____.___ O mg/L O %	Flag
pH	Cal. STD 1 Description _____	Cal. STD 1 Value ___.___	Cal. STD 2 Description _____	Cal. STD 2 Value ___.___ Flag
CONDUCTIVITY	Cal. STD 1 Description _____	Cal. STD 1 Value _____	Cal. STD 2 Description _____	Cal. STD 2 Value _____ Flag

QUALITY CONTROL CHECK (Perform at least once per week)

TIME: ___:___

QC BATCH #: _____

Date Prepared: _____

O No QC Check performed at this visit?

Parameter	TEMP. (°C)	COND (µS)	pH	FLAG
Standard:				
Measured:				

COMMENTS: _____

POST-MEASUREMENT CALIBRATION CHECK

	pH	COND (µS)	FLAG
Standard:			
Measured:			

SECCHI DEPTH

TIME: ___:___

Secchi Depth (m) XX.X:

	DISAPPEARS:	REAPPEARS:
Reading 1:		
Reading 2:		
Reading 3:		

CLEAR TO BOTTOM? O Yes O No

SECCHI FLAG: _____

Flag	Comments	Unique Flag and Comments entered here are for both sides of this Field Measurement form.

Flag codes: K = Sample not collected; U = Suspect sample; F1, F2, etc. = flag assigned by field crew.

9117512028

NCCA 2010 FIELD MEASUREMENT (Back)

Reviewed by (initial): _____

SITE ID: **NCCA10-** _____ DATE: ___/___/2010

Hydrographic Profile

Intervals (m): 0.1m below surface, 0.5 below the surface, every 1 meter from depths of 1.0 to 10m, and every 5 meters thereafter if the site is greater than 10m. Take the last set of measurements at 0.5m from the bottom.

STATION DEPTH (m): _____ XX.X

Complete either SAL or COND:

	DEPTH(m) XX.X	TEMP. (°C) XX.X	pH XX.XX	DO(mg/L) XX.X	SAL (‰) (Marine) XX.X	COND (uS) (Great Lakes) XXX.X	LIGHT(AMB) XXX.X	LIGHT(UW) XXX.X	FLAG
DOWN CAST	0.1								
	0.5								
Near Bottom									
UP CAST									

Flag codes: K = Sample not collected; U = Suspect sample; F1, F2, etc. = flag assigned by field crew.
Record any Profile flag and comments on front side of this field measurement form.

4981512022

NCCA 2010 SAMPLE COLLECTION FORM - (Front)

Reviewed by (Initials): _____

SITE ID: NCCA10- DATE: ___/___/2010

WATER CHEMISTRY , CHLOROPHYLL and NUTRIENT COLLECTION (0.5m)

Water Chemistry (Non-Filtered)	Chilled	Comments	No Sample Collected
	○		

Chlorophyll-a	Frozen	Vol Filtered (ml)	Comments	No Sample Collected
	○			

Nutrients (Filtered)	Chilled	Comments	No Sample Collected
	○		

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

ENTEROCOCCI (Target Volume = 250 mL)

No Sample Collected ○

Sample ID (One unique ID per line)		Time Collected (hhmm)	Depth Collected (m)	Sample Volume (mL)	Filt. Start Time (hhmm)	Volume Filtered (Target = 50 mL)				Vol. of Rinse	Filt. End Time (hhmm)	Time Frozen (hhmm)	Flag
						Filt. 1	Filt. 2	Filt. 3	Filt. 4				
Filter Blank						Volume Filtered (Target = 20 mL)							
	○ F					Filt. 1	Filt. 2	Filt. 3	Filt. 4				

Flag codes: K = No measurement made, U = Suspect measurement., F1,F2, etc. = flags assigned by each field crew. Explain all flags in comments.

Flag	Comments

GREAT LAKES ONLY

PHYTOPLANKTON (Amber Nalgene 1-L)

No Sample Collected ○

Sample ID	Preserved	Depth Collected (m)	Time Collected (hhmm)	Time Preserved (hhmm)	Comments
	○				

Under water video camera Digital Video Recording

No Sample Collected ○

File name Format: DVRyymmdd_hhmm_xxx.avi				Transferred to SD Card	Comments
DVR			.avi	○	

NCCA 2010 SAMPLE COLLECTION FORM - (Back)

Reviewed by (initial): _____

SITE ID: NCCA10-_____ DATE: ___/___/2010

BENTHIC INFAUNA COLLECTION

No Sample Collected ○

GRAB AREA (m²): _____ GRAB TYPE: ○ Van Veen ○ Standard Ponar ○ Other: _____

NOTE:

SIEVE SIZE: ○ 0.5 mm ○ 1.0 mm NUMBER OF GRABS: ○ 1 ○ 2 2 Grabs are required for samplers less than 0.03 m²

Sample ID	Depth (cm) (Must be >7 cm)	No. of Jars	Preserved	Comments
			○	

SEDIMENT CHARACTERISTICS (Benthic Grab)

COLOR: ○ Black ○ Brown ○ Light Brown ○ Dark Brown ○ Gray ○ Other _____

SUBSTRATE: ○ Sand ○ Muck ○ Gravel ○ Cobble ○ Shellhash ○ Other _____

SMELL: ○ Fishy ○ Chemical ○ Sulphur ○ None ○ Other _____

SURFACE: ○ Film ○ Floc ○ Nothing Noted ○ Other _____

VISIBLE FAUNA: ○ Yes ○ No TYPE: _____

VISIBLE FLORA: ○ Yes ○ No TYPE: _____

SEDIMENT SAMPLE COLLECTION

DISTANCE SEDIMENT COLLECTED:

○ Within 37m from X-site ○ Between 37-100m from X-site ○ Between 100-500m from X-site (GREAT LAKES ONLY)

SEDIMENT ORGANICS/METALS (Glass Jar 500 ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 250 mL)	Chilled	Comments
		○	

SEDIMENT GRAIN SIZE (Nalgene 125ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 100 mL)	Chilled	Comments
		○	

SEDIMENT TOC (Nalgene 60ml)

No Sample Collected ○

Sample ID	Sample Volume (Target = 50 mL)	Chilled	Comments
		○	

SEDIMENT TOXICITY (1 gal Screw Top Bucket)

No Sample Collected ○

Sample ID	Sample Volume (Target = 3L)	Chilled	Comments
		○	

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

NCCA 2010 ECO FISH COLLECTION (Front)

Reviewed by (initial): _____

SITE ID: NCCA10- _____ DATE: ___/___/2010

Collection Method

- ○ TRAWL
- ○ HOOK & LINE
- ○ SEINE
- ○ GILL NET
- ○ PURCHASED DOCKSIDE
- ○ OTHER EXPLAIN: _____

Trawl/Seine Info: HELMSMAN: _____ LINE OUT (m): _____

Trawl/Seine Start:

HEADING IN DEGREES MAGNETIC: _____

	LATITUDE:	LONGITUDE:	START TIME:
Decimal Degrees	___ . _____	___ . _____	___ : ___

FLAG: _____

Trawl/Seine End:

	LATITUDE:	LONGITUDE:	END TIME:
Decimal Degrees	___ . _____	___ . _____	___ : ___

FLAG: _____

If Seine: LENGTH: _____ (m) FLAG: _____

If Gill Net: LENGTH: _____ (m) START TIME: ___ : ___ END TIME: ___ : ___ FLAG: _____

Flag Codes: K = no measurement made; U = suspect measurement; F1, F2, etc. = flags assigned by each field crew. Explain all flags in comments section.

Flag	Comments

NCCA 2010 ECO FISH COLLECTION (Back)

Reviewed by (initial): _____

SITE ID: NCCA10- DATE: ___ / ___ / 2 0 1 0 PAGE: ___ OF ___

FISH TISSUE SAMPLE

○ NO SAMPLE COLLECTED

SAMPLE ID _____

FISH ALL WITHIN 75% OF LARGEST SPECIMEN ○

FISH ARE ALL THE SAME SPECIES ○

	Genus Species	Total Length (mm)	Frozen	Comments
.01			○	
.02			○	
.03			○	
.04			○	
.05			○	
.06			○	
.07			○	
.08			○	
.09			○	
.10			○	
.11			○	
.12			○	
.13			○	
.14			○	
.15			○	
.16			○	
.17			○	
.18			○	
.19			○	
.20			○	

The eco fish composite must consist of at least 5 fish of adequate size to provide a total weight of 500 grams of whole-body tissue.

NCCA 2010 HUMAN HEALTH FISH COLLECTION (Front) Reviewed by (initial): _____

SITE ID: NCCAGL10- DATE: ___/___/2010

Collection Method

○ TRAWL ○ HOOK & LINE ○ SEINE
○ GILL NET
○ OTHER EXPLAIN:

Trawl/Seine Info: HELMSMAN: LINE OUT (m):

Trawl/Seine Start: HEADING IN DEGREES MAGNETIC:

Decimal Degrees LATITUDE: . LONGITUDE: . START TIME: :

FLAG:

Trawl/Seine End:

Decimal Degrees LATITUDE: . LONGITUDE: . END TIME: :

FLAG:

If Seine: LENGTH: (m) FLAG:

If Gill Net: LENGTH: (m) START TIME: : END TIME: : FLAG:

Flag Codes: K = no measurement made; U = suspect measurement; F1, F2, etc. = flags assigned by each field crew. Explain all flags in comments section.

Flag	Comments

NCCA 2010 HUMAN HEALTH FISH COLLECTION (Back)

Reviewed by (initial): _____

SITE ID: NCCAGL10- DATE: ___ / ___ / 2010 PAGE: ___ OF ___

FISH TISSUE SAMPLE

○ NO SAMPLE COLLECTED

SAMPLE ID _____

FISH ALL WITHIN 75% OF LARGEST SPECIMEN ○

FISH ARE ALL THE SAME SPECIES ○

	Genus Species	Total Length (mm)	Frozen	Comments
.01			○	
.02			○	
.03			○	
.04			○	
.05			○	
			○	
			○	
			○	
			○	
			○	

Human health fish composites should consist of 5 similarly-sized adult fish of the same species that will collectively yield about 500 g of fillet tissue (refer to Frequently Asked Questions for possible exceptions).

NCCA 2010 SITE ASSESSMENT (Front)

Reviewed by (initial): _____

SITE ID: NCCA10- _____ DATE: ___/___/2010

SHORELINE ACTIVITIES AND DISTURBANCES OBSERVED (Intensity: Blank=Not observed, L=Low, M=Moderate, H=Heavy)

Residential	Recreational	Agricultural	Industrial	Management
L M H Residences	L M H Hiking Trails	L M H Cropland	L M H Industrial Plants	L M H Chemical Treatment
L M H Maintained Lawns	L M H Parks, Campgrounds	L M H Pasture	L M H Mines/Quarries	L M H Angling Pressure
L M H Construction	L M H Primitive Parks, Camping	L M H Livestock Use	L M H Oil/Gas Wells	L M H Dredging
L M H Pipes, Drains	L M H Trash/Litter	L M H Orchards	L M H Power Plants	L M H Channelization
L M H Dumping	L M H Surface Films	L M H Poultry	L M H Logging	L M H Water Level Fluctuations
L M H Roads	L M H Dunes	L M H Irrigation Equip.	L M H Evidence of Fire	L M H Shoreline Hardening
L M H Bridge/Culverts	L M H Beach	L M H Water Withdrawal	L M H Odors	L M H Dredge Material
L M H Sewage Treatment	L M H Forested		L M H Commercial	

SITE CHARACTERISTICS (200 m radius)

Waterbody Character	Pristine	O 5	O 4	O 3	O 2	O 1	Highly Disturbed
	Appealing	O 5	O 4	O 3	O 2	O 1	Unappealing
Dominant Land Use	Dominant Land Use Shoreline	O Forest	O Agriculture	O Range	O Urban	O Suburban/Town	
	If Forest, Dominant Age Class	O 0 - 25 yrs.	O 25 - 75 yrs.	O > 75 yrs.			

WEATHER

GENERAL ASSESSMENT (Biotic integrity, Vegetation diversity, Local anecdotal information)

NCCA 2010 SITE ASSESSMENT (Back)

Reviewed by (initial): ____

SITE ID: NCCA10- DATE: ___/___/2010

GENERAL ASSESSMENT (continued)

APPENDIX C

Example of Great Lakes Disinfection Protocols from Wisconsin DNR

Boat and Gear Disinfection Protocol

Boat and trailer cleaning guidelines to prevent the spread of aquatic invasive species have been widely distributed to the public through a variety of publications, pamphlets, signs, etc. The guidelines consist of a nationally-accepted set of prevention steps. While disinfection is **not** a required prevention step for the general public, some boaters may be interested in the disinfection procedures followed by the WI DNR. Please note: the first three steps (Inspect and Remove, Drain, and Dispose) listed below are required.

The following steps shall be taken every time a boat, equipment or gear is moved between waters to avoid transporting invasive species and/or pathogens:

1. **Inspect** and **remove** aquatic plants, animals, and mud from your boat, trailer, equipment and gear.
2. **Drain** all water from your boat, motor, live well, bilge, transom wells, as well as from your equipment and gear, including but not limited to tracked vehicles, barges, silt or turbidity curtain, hoses, sheet pile and pumps.
3. **Dispose** of unwanted aquatic plants and animals in an appropriate way.
4. **Disinfect** your boat, equipment and gear by either:
    - **Washing** with ~212º F water (steam clean), OR
    - **Drying** thoroughly for 5 days after cleaning with soap and water and/or high pressure water, OR
    - **Disinfecting** with either 200 ppm (0.5 oz per gallon or 1 Tablespoon per gallon) Chlorine for 10 minute contact time or 1:100 solution (38 grams per gallon) of Virkon Aquatic for 20 to 30 minute contact time. Note: Virkon is not registered to kill zebra mussel veligers nor invertebrates like spiny water flea. Therefore this disinfect should be used in conjunction with a hot water (>104º F) application.

Safety Precautions for Disinfectant Use

Virkon-A:

1. Receive and be required to read a copy of the Virkon-A Materials Safety Data Sheet (MSDS) for the product.
2. Wear chemical splash goggles.
3. Wear a face shield where the possibility exists for face contact due to splashing or spraying of the material.
4. Wear impervious clothing to prevent contact with skin. (gloves, pants, jacket, hood, and boots) or a Tyvek style full body suit.

    In addition, all employees who handle or mix Virkon-A in powder form and prefer to wear a dust mask respirator when handling powder, may do so in compliance with the DNR Respiratory Protection Program Handbook MC 9180.5 Voluntary Use requirements.

Bleach:

Follow precautions 2, 3, and 4 (above).

- Chlorine Wear eye protection, rain gear, gloves if spraying. Stay upwind of the spray. Will break down in sunlight and when in contact with organic material. Is corrosive to metal and rubber. Is toxic to fish at these concentrations so rinse

well after disinfection or neutralize with sodium thiosulfate. For neutralizing chlorine, spray sodium thiosulfate in an 800 ppm solution (3 grams per gallon of water) on all surfaces after the disinfection period is over. Rinse with water from the next lake to remove any remaining sodium thiosulfate.

- Virkon Aquatic This is a disinfectant in the peroxygen (hydrogen peroxide) family. It is a powder. It is 99.9% biodegradable and breaks down to water and oxygen and is not corrosive at the working dilution. Wear dust mask if mixing powder and eye protection, rain gear and gloves if spraying. Stay upwind of spray.

Sources of disinfectants

Chlorine - Household bleach (5.25% chlorine) can be purchased from a grocery or convenience store. HTH is granular chlorine (70% calcium hypochlorite) and can be purchased from a pool supply company.

Sodium Thiosulfate - Commonly used to neutralize chlorine and iodine. It should be available at a pool supply company or from a chemical supply company.

Virkon Aquatic is available from Western Chemical. It is the same formulation, but without the perfume and dye, and the label addresses specific fish pathogens. Their phone is 1-800-283-5292.

Disinfection measures must be taken prior to moving boats, equipment and other gear from one waterbody to another. They are not needed daily when sampling the same waterbody or for law enforcement equipment in emergency situations. In cases where boats and gear return to state hatcheries, disinfection should be done in a location away from ponds and water supplies to prevent disinfectant or untreated water from entering those areas. Every effort should be made to keep the disinfection solution and rinse water out of surface waters.
To the extent practicable, equipment and gear used on waters known to be infested with invasive species and viruses should not be used on other non-infested waters. The following are some helpful hints to consider when planning your work in water.

- Organize your sampling so the work in infested waters is always done last.

- If a high percentage of your work is done in waters with invasive species, consider dedicating certain gear to be used only in those waters.

- Depending on the type of work you are doing, it may be possible to work with lake volunteers and use their boats to collect samples. That way only your gear needs to be disinfected.

The following methods are provided to assist staff when disinfecting equipment and gear commonly used by department staff.

Nets
Organic debris should be removed prior to disinfection. Power washing is not required, but nets could be sprayed with a garden hose to remove debris. Nets may be steam cleaned, washed and dried thoroughly for five days or treated with a disinfection solution. Nets should be placed

in the disinfection solution for the appropriate contact time for the solution being used. After rinsing, the nets can be used immediately, or hung to dry.

Personal protective gear, including rain gear, gloves, boots/waders
Scrub personal protective gear with the disinfection solution. After scrubbing, the gear should be kept wet with the disinfection solution for the appropriate contact time. Rinse with clean water or water from the next waterbody. Alternatively, personal gear may be steam cleaned or dried thoroughly for five days after cleaning with soap and water.

Dip nets, measuring boards and other sampling gear
Remove any organic material from sampling gear. There are several options for disinfecting smaller gear. *Dissolved oxygen probes and other sensitive electronic sampling gear may be damaged by disinfection solution and should only be rinsed with clean water*. For other gear used in water choose one of the following options:

- Option one: The gear can be sprayed with the disinfection solution and a wet surface maintained for the appropriate contact time. The gear should be rinsed with clean water or water from the next waterbody before it is used again.
- Option two: Fill a tub with disinfection solution and place all equipment in the tub for the appropriate contact time. The gear should be rinsed with clean water or water from the next waterbody before it is used again.
- Option three: Use a completely new set of gear for each waterbody during the work day and disinfect all gear at the end of the day using option one or two.

Boats, trailers, and live wells
Remove organic material from boats, trailers, and live wells. Drain water from live wells, bilges and pumps. The outside and inside of the boat, trailer, live wells, bilges, and pumps should be sprayed with the disinfection solution and left wet for the appropriate contact time. The inside of the live wells, bilges and pumps should be made to contact the solution for the appropriate contact time as well. Run pumps so they take in the disinfection solution and make sure that the solution comes in contact with all parts of the pump and hose. The boat, trailer, bilges, live well, and pumps should be rinsed with clean water or water from the next waterbody after the appropriate contact time. *Every effort should be make to keep the disinfection solution and rinse water out of surface waters.* Pull the boat and trailer off the ramp and onto a fairly level area and away from street drains to minimize potential runoff into surface waters.

Motors
After removing from the water, tip the motor to the down position and start the motor for several seconds or turn motor over several times to dispel water from the cooling system. Alternatively and especially for motors moored in water for several days or more, emerge the lower unit in a bucket of disinfectant and run the motor to ensure contact with all internal parts and allow for the appropriate contact time. Or, rig up a short (6 foot) piece of garden hose to lower unit muffs. A pail of the disinfectant can be set in the back of the boat and gravity fed to the lower unit to run the disinfectant through the motor. Allow solution to remain in motor for the appropriate contact time. The hose will need to be primed to start the gravity flow because the lower unit does not create enough suction to prime the hose. A non-corrosive (Virkon Aquatic) is recommended for use to protect the impeller. Rinse with clean water or water from the next waterbody.

Heavy Equipment
For heavy equipment steam-cleaning is an effective method of disinfection.

APPENDIX D

Shipping and Tracking Guidelines

Tracking Forms

If you have access to a computer, fill out the **electronic tracking forms**:

- Be careful to fill out all information accurately and completely.
- If you do not have a printer, you will need to fill out and include the scannable form in the shipping cooler. This is true for all tracking forms listed below.

4 Tracking Forms

1 - Tracking and Sample Status – WRS

- This form is filled out for the samples that are shipped immediately after each sampling event (water chemistry, chlorophyll-*a*, sediment grain size, sediment TOC, and the dissolved nutrients samples).
- All of these samples will be sent together in one cooler to the EPA Corvallis lab.
- Save form according to the file naming convention on the bottom of form.
- Email to address on bottom of form and print form to include in the shipping cooler.

**Emailing the electronic WRS form serves as the "status report" for that sampling event.*

2 - Tracking (Batched Samples)

- BATCHED samples are held & shipped within 2 weeks of collection. Send form when SHIPPED.
- Use one tracking form for each laboratory.
- Save form according to the file naming convention on the bottom of form.
- Email to address on bottom of form and print form to include in the shipping cooler.

3 - Tracking – Fish Tissue

- Fish for ecological tissue analyses (eco fish) will be collected at all sites.
- A separate tracking form is provided for eco fish.
- The eco fish will be sent to the eco fish tissue lab.
- Save form according to the file naming convention on the bottom of form.
- Email to address on bottom of form and print form to include in the shipping cooler.

4 – Tracking – Human Health Fish Tissue

- A subset of 150 Great Lakes sites will be sampled for human health contaminants in fish tissue.
- A separate tracking form is provided for human health fish.
- The human health fish will be sent to the human health fish tissue lab.
- Save form according to the file naming convention on the bottom of form.
- Email to address on bottom of form and print form to include in the shipping cooler.

If you cannot use a computer before shipping:

- Fill out the paper version of the tracking form.
- Notify the Information Management Center (contact info on bottom of form) – either FAX form or leave voice message with ALL info from the form.
- Include the form in the shipping cooler.
- Make sure to FAX or leave a voice message BEFORE the form is sealed in the cooler!

Status Report

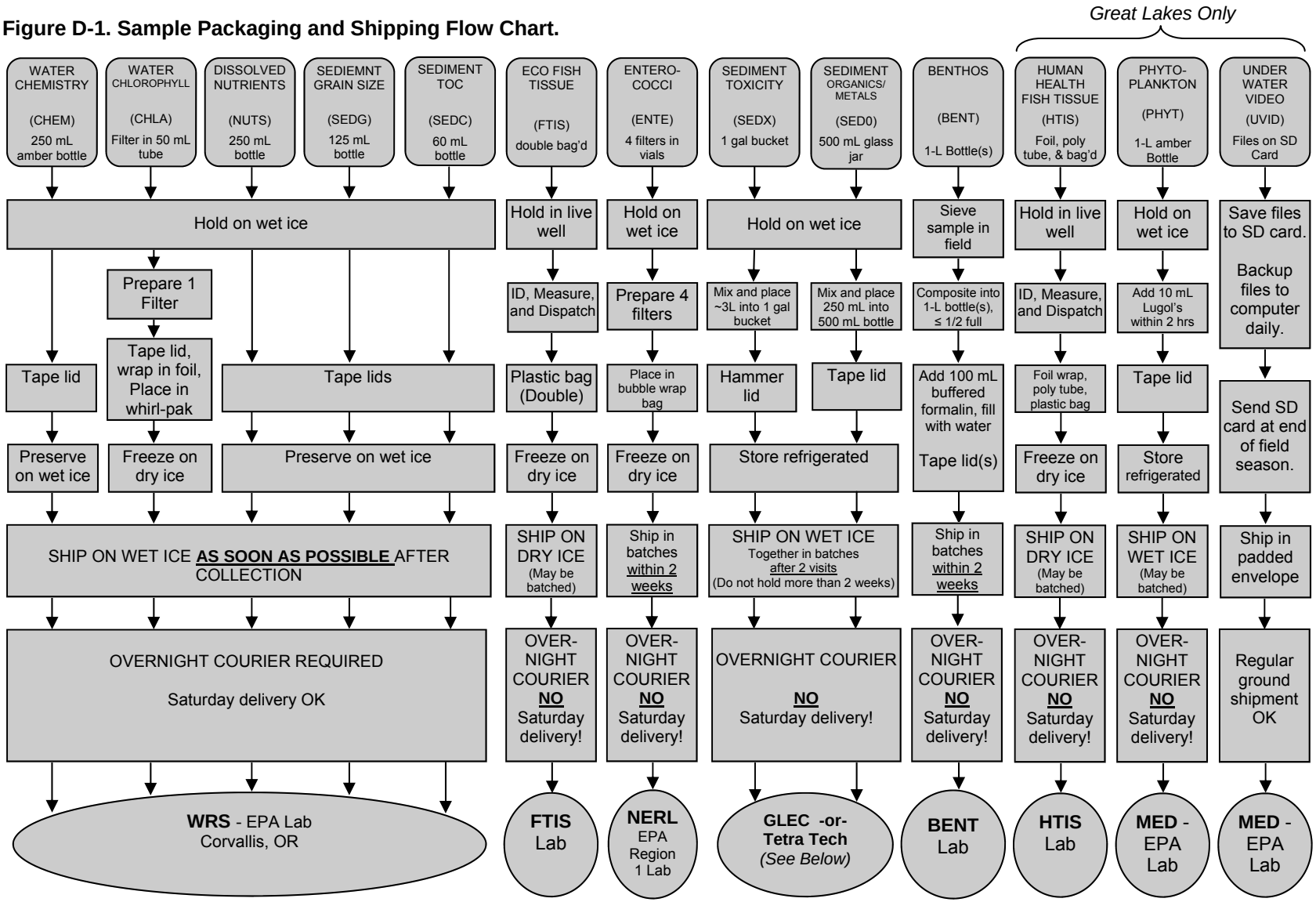
- After each site, the Field Team Leader must file a status report with the Information Management Center and the Field Logistics Coordinator to track visits/samples and to describe activities, problems, and requests.
- Emailing the electronic WRS form serves as the status report and will go to both the Information Management Center and the Field Logistics Coordinator.
- If the form cannot be emailed, faxing or phoning the information serves as the status report.

SHIPPING GUIDELINES

Before shipping, it is very important to preserve each sample as directed in the sample collection portion of this Field Operations Manual.

- Preserve the samples as specified for each indicator before shipping (Figure D-1).
- Be aware of the holding times for each type of sample (Table D-1):
- Water chemistry, dissolved nutrients, sediment grain size and sediment TOC samples must be shipped within 24 hours of collection.
- Chlorophyll-*a* samples have a longer holding time, but will be sent with the water chemistry samples since they are analyzed by the same laboratory.
- The benthos samples must be preserved immediately upon collection; they may then be sent in batches to the appropriate laboratory.
- The sediment toxicity and sediment chemistry (organics/metals) samples have a two week holding time. These two sample types will be shipped to the same sediment lab, but no more than two visits worth of samples will be able to fit in a single cooler. The extra space within the cooler will allow for sufficient wet ice for shipping. Securely tape the cooler drainage open to prevent pressure build-up in the cooler.
- Secure the cooler with strapping tape.
- Enterococci samples must be filtered and frozen on dry ice within 6 hours of collection. They can be batched and sent every two weeks.
- Fish tissue samples must be frozen on dry ice as soon as possible (hold on wet ice until freezing on dry ice).
- See “Dry Ice Shipping Protocols” at the end of this Appendix.

Figure D-1. Sample Packaging and Shipping Flow Chart.



Field Forms (PACK): All field forms should be reviewed and sent in to the Information Management Coordinator (WED – Corvallis) every 2 weeks

Sediment Toxicity (SEDX) and **Sediment Organics/Metals (SEDO)** will be shipped together to either Tetra Tech or GLEC based on the state in which the sampling took place. Refer to the note enclosed with the FedEx airbills to determine the correct destination lab.

WATER CHEMISTRY, CHLOROPHYLL-*a* and DISSOLVED NUTRIENTS

- **Water Chemistry**
  Stored in a 250mL amber Nalgene bottle
  - Confirm that the labels with sample IDs are completed and covered with clear tape.
  - Seal the lid with plastic (electrical) tape.
  - Place the bottle in a 1 qt self-sealing plastic bag and place inside the cooler liner.
- **Chlorophyll-*a***
  Filter stored in a 50 mL screw-top centrifuge tube wrapped with aluminum foil
  - Confirm that the labels with sample IDs are completed and covered with clear tape.
  - Seal the lid with plastic (electrical) tape.
  - Cover the tube with aluminum foil.
  - Place the centrifuge tube in a whirl-pak.
  - Place the whirl-pak in a small (sandwich size) self-sealing plastic bag and place inside cooler liner with water chemistry sample.
- **Dissolved Nutrients Sample**
  Filtrate from chlorophyll-*a* filtration collected in a 250 mL Nalgene bottle.
  - Confirm that the labels with sample IDs are completed and covered with clear tape.
  - Seal the lid with plastic (electrical) tape.
  - Place the bottle in a 1 qt self-sealing plastic bag and place inside cooler liner with water chemistry sample..

SEDIMENT GRAIN SIZE AND TOC SAMPLES
Stored in 125 mL bottles (grain size) and 60 mL bottles (TOC).
- Confirm that the labels with sample ID is completed and covered with clear tape.
- Seal the lids with plastic (electrical) tape.
- Place each of the bottles in separate small (sandwich size) self-sealing plastic bags.
- Place the bags in a 1 gal self-sealing plastic bag and place inside cooler liner with water chemistry sample.

SEDIMENT TOXICITY AND ORGANICS/METALS SAMPLES
Stored in 1 gallon screw-top bucket (toxicity) and 500 mL glass jar (organics/metals).
- Confirm that the labels with sample ID is completed and covered with clear tape.
- Seal the lids of glass jars with plastic (electrical) tape.
- Hammer the lids of plastic buckets in clockwise direction to close securely
- Wrap each of the 500 mL glass jar in bubble wrap and place in separate 1 qt self-sealing plastic bags.
- Place the bags in a 1 gal self-sealing plastic bag and place inside cooler liner with sediment toxicity samples
- These two sample types will go to the same sediment lab, but no more than two visits worth of samples (2 buckets and 2 glass jars) will be able to fit in a single cooler. This is to allow for enough space for wet ice in the cooler.
- Do not hold these samples longer than 2 weeks.

UNDERWATER VIDEO RECORDING FILES (GREAT LAKES SITES ONLY)
One minute of good quality video is recorded on the system DVR at the sampling site.
- Transfer the files to the SD card provided daily.
- Backup the video file daily or as often as possible to a computer hard drive.
- Send SD card to MED lab in Duluth, MN at end of field season.

PHYTOPLANKTON (GREAT LAKES SITES ONLY)

Stored in a 1 L amber Nalgene bottle

- Confirm that the labels with sample IDs are completed and covered with clear tape.
- Add 10 mL Lugol's solution with 2 hours of sample collection
- Seal the lid with plastic (electrical) tape.
- Place the bottle in a 1 gal self-sealing plastic bag and place inside a cooler with liner.
- Shipping with wet ice is preferred, but not required for these samples.
- Samples can be held for up to 2 weeks and shipped in batches to the laboratory for analysis.

BENTHIC INVERTEBRATE SAMPLES

Preserved in 100 % buffered formalin and sealed at the site with plastic (electrical) tape.

- Confirm that the label with sample ID is completed and covered with clear tape.
- Check to make sure jars are sealed with electrical tape.
- Place up to ten 1 L jars in each cooler.
- Surround the jars with crumpled newspaper, vermiculite, or other absorbent material.
- Samples can be held for up to 2 weeks and shipped in batches to the laboratory for analysis.

FISH TISSUE SAMPLES

The samples need to be frozen as soon as possible after collection (within 6 hours).

- Pack the cooler with 50 lbs of dry ice.
- Refer to the DRY ICE SHIPPING PROTOCOLS at the end of this Appendix.
- Samples may be stored on dry ice for a maximum of 24 hours. Sampling teams have the option, depending on site logistics, of:
    - shipping the samples packed on dry ice (50 pounds, layered to ensure direct contact between fish and dry ice), via priority overnight delivery so that they arrive at the sample preparation laboratory within 24 hours of sample collection, or
    - freezing the samples within 24 hours of collection at ≤-20°C, and storing the frozen samples until shipment within 2 weeks of sample collection (frozen samples will be packed on layered dry ice and shipped to the sample preparation laboratory via priority overnight delivery service).

ENTEROCOCCI SAMPLES

The sample needs to be filtered and frozen as soon as possible after collection (within 6 hours).

- 4 filters are placed in separate microcentrifuge tubes with ceramic beads
- Confirm that the tubes are properly sealed and labeled with the volume of sample filtered (*do not cover the small labels on the microcentrifuge tubes with tape*).
- Place the 4 microcentrifuge tubes in the bubble wrap envelope.
- Confirm that the bubble wrap envelope is labeled with the appropriate sample ID and volume of sample filtered. Covered outer label with clear plastic tape.
- Place the bubble wrap envelope in the cooler and close.
- Pack the cooler with 10-15 lbs of dry ice (10 lbs if using dry ice blocks or slices, 15 lbs if using dry ice pellets).
- Refer to the DRY ICE SHIPPING PROTOCOLS at the end of this Appendix.
- Samples can be held frozen and shipped in batches to the laboratory for analysis.
- Do not hold these samples longer than 2 weeks.

DRY ICE SHIPPING PROTOCOLS

1. Indicate dry ice on shipping airbill

    - Fill out Section 1 and Section 3 of the Fed Ex airbill with your Sender and Recipient address and phone number.
    - In Section 4, check "FedEx Priority Overnight."
    - In Section 5, check "Other."
    - In Section 6, under "Does this shipment contain dangerous goods?":
        - Check "Yes/Shipper's Declaration not required."
        - Check "Dry Ice," and fill out "_1_ x _(*amt. of dry ice in kg*)_ kg"
    - In Section 7, fill out weight and declared value of package.

2. Label cooler with a Class 9 Dangerous Goods label (available from FedEx) (Fig. D-2).

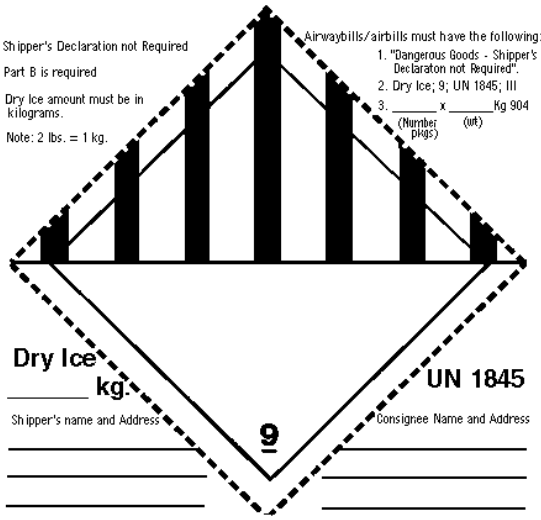


Figure D-2. Class 9 Dangerous Goods label.

- Place the label on the front side of the cooler, not the top of the cooler.
- Fill out #3 in the top right hand corner of the label with the same information as in Section 6 of the FedEx airbill.
- Declare the weight of the dry ice again in the lower left hand corner.
- Fill out the Sender ("Shipper") and Recipient ("Consignee") address on the bottom of the label.

3. Securely tape the cooler drainage open to prevent pressure build-up in the cooler. This is critical to ensure proper venting of the dry ice.
4. Secure the cooler with strapping tape.
5. Place the completed airbill on the top of the cooler.

NOTE: *Not all FedEx locations will accept shipments containing dry ice. Dry ice shipments can be shipped from "FedEx staffed" locations. You can also arrange for a pick-up from your lab or hotel. Dry ice shipments usually cannot be shipped from FedEx Kinko's Office and Print Centers® or FedEx Authorized ShipCenter® locations. These types of locations are differentiated on FedEx.com in the "Find FedEx Locations" feature. Please be sure to call in advance to ensure your location will accept the package for shipment.*

TRACKING FORMS

A Tracking Form must be filled out to accompany each sample shipment. Please refer to Figures 3-2 through 3-5 for examples of Tracking Forms completed for both unpreserved and

preserved samples. Be very careful to fill in the information correctly and legibly, especially the airbill number, Site ID, and Sample ID numbers. Use the codes on the bottom of the form to indicate sample type. Fill out a separate tracking form for each destination laboratory.

The Tracking Form is to be placed in a self-sealing plastic bag and included inside the shipping container. Before sealing the container, remember to submit the status report (via email) to sampletracking@epa.gov (see Section 3.2.5); you will need the information on the tracking form to fill out the status report form. For preserved samples, submit a tracking form both when the samples are brought to the holding facility AND when they are shipped to the appropriate laboratory. For each shipment, you must print the electronic form or fill out a scannable tracking form to include in the cooler and submit the electronic status report.

When ice is used for shipment (water chemistry, dissolved nutrients, chlorophyll-*a*, sediment grain size and TOC):

- Ensure that the ice is fresh before shipment; pack the entire cooler full with ice.
- Line the cooler with a large, 30 gallon plastic bag.
- Contain the ice separately within numerous 1 gallon self-sealing plastic bags. Double-bag the ice.
- Use white or clear bags and label with a dark indelible marker. Label all bags of ice as “ICE” to prevent misidentification by couriers of any water leakage as a possible hazardous material spill.
- Place bagged samples and bags of ice inside the cooler liner and seal the liner.
- Secure the cooler with strapping tape.

When dry ice is used for shipping (fecal indicator and fish tissue):

- Indicate dry ice on shipping airbill.
- Label cooler with a Class 9 Dangerous Goods label.

Table D-1 Sample Packaging and Shipping Summary

SAMPLE TYPE		SAMPLE TARGET VOLUME	CONTAINER	PRESERVATIVE	SHIPPING TIME FRAME	PACKAGING FOR SHIPMENT
Water Chemistry		250 mL	250 mL amber Nalgene bottle	Wet ice in field	Immediate	Ship in cooler with wet ice
Chlorophyll-*a*	Collection	2 L	2L amber Nalgene bottle	Wet ice in field		
	Processing	Readily visible stain in filter - max of 2000 mL filtration	Filter in 50 mL centrifuge tube	Dry ice in field after filtration	Immediate	Ship in cooler with wet ice
Dissolved Nutrients		250 mL of filtrate from chl-*a* filtering	250 mL Nalgene bottle	Wet ice in field	Immediate	Ship in cooler with wet ice
Sediment - Organics/Metals		250 mL	500 mL glass jar	Wet ice in field; refrigerate to hold	Batch up to 2 weeks	Ship in cooler with wet ice
Sediment - Grain size		100 mL	125 mL Nalgene bottle	Wet ice in field	Immediate	Ship in cooler with wet ice
Sediment - TOC		50 mL	60 mL Nalgene	Wet ice in field	Immediate	Ship in cooler with wet ice
Sediment - Toxicity		3 L	1 gallon screw top bucket	Wet ice in field; refrigerate to hold	Batch up to 2 weeks	Ship in cooler with wet ice
Benthic macroinvertebrates		All organisms in grab(s)	1 L Nalgene bottle(s)	100 mL 100% buffered formalin, fill to rim with water	Batch up to 2 weeks	Ship in cooler or sturdy container
Enterococci	Collection	250 mL	Pre-sterilized 250 mL Nalgene bottle	Wet ice in field		
	Processing	4 - 50 mL filtrations	4 filters in microcentrifuge tubes	Dry ice in field; hold in freezer; MUST be filtered & frozen within 6 hours of collection	Batch up to 2 weeks	Ship in cardboard cooler box (provided) with DRY ICE
	Filter Blanks (Revisit sites)	4 - 20 mL filtrations of sterile saline	4 filters in microcentrifuge tubes	Dry ice in field; hold in freezer	Batch up to 2 weeks, send with regular ENTE sample from site	Ship in cardboard cooler box (provided) with DRY ICE
Eco Fish Tissue		5 – 20+ fish (500 g whole-body tissue)	2 gallon self-sealing bag(s) Large outer plastic bag	Dry ice in field; hold in freezer	Batch up to 2 weeks	Ship in cooler with DRY ICE
Phtyoplankton (Great Lakes Only)		1 L	1 L amber Nalgene bottle	10 mL Lugols solution within 2 hrs Wet ice in field; refrigerate to hold	Batch up to 2 weeks	Ship in cooler with wet ice (preferred but not required)
Underwater Video (Great Lakes Only)		1 minute video	Save to DVR	Save files to SD card daily Backup to computer often	Ship SD card at end of field season	Ship in padded envelope
Human Health Fish Tissue (Subset of Great Lakes sites)		5 Fish (500 g of fillet tissue)	Wrapped individually in solvent rinsed foil Sealed in poly tubing Large outer plastic bag	Dry ice in field; hold in freezer	Batch up to 2 weeks	Ship in cooler with DRY ICE