
WATER QUALITY PROGRAMS DIVISION

Standard Operating Procedure for the Collection of Fish for
Toxics Analysis

Adopted May 2005

Draft Copy



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STANDARD OPERATING PROCEDURE FOR THE COLLECTION OF FISH FOR TOXICS ANALYSIS ADOPTED MAY 2005

1.0 General Information

Collection of fish for toxics analysis is an integral component of special studies involving pesticides, other organics, or heavy metals. The OWRB does not have the regulatory capacity to issue fish consumption advisories. Therefore this document is not meant to provide guidance for the collection of fish for this purpose. Fish collections for the purposes of toxic analysis as related to this SOP are strictly to determine the potential bioaccumulation of organics and heavy metals on certain fish populations in areas involved in special studies or of special concern. The analytical data is forwarded to the ODEQ for further processing.

Following is a detailed description of sampling procedures. Efficiency is the key, and finding a comfortable sequence of sampling is essential. This will vary from person to person and from sampling team to sampling team. Yet, employing consistent sampling patterns at every site will maximize the number of sites sampled per day and decrease the chance for introduction of sampling error.

2.0 Definitions/Terms

- Team Leader—crew member of fish collection team who provides support, expertise, and opinions; gives instruction and has final say on how work will be done; must score a 95% on critical fish identification
- Team Member—crew member of fish collection team who provides support, expertise, and opinions; follows the instructions of the team leader
- Hoop Netting—placement of various sized nets in medium to deep water; selective for areas of instream cover as well as larger fish with more surface area
- Electrofishing—use of an electrical field in the water to stun fish for collection; selective for areas of instream cover as well as larger fish with more surface area
- Seining—wide and deep net dragged through water to collect fish; selective for runs and pools as well as smaller fish with less surface area
- Gill Netting—place of block net of various sized mesh in deeper waters. Fish become trapped in the net and are harvested after a period of time.

3.0 Safety

Upon reaching the sampling location, site safety determinations should be made before proceeding. Please refer to the OWRB safety manual for instructions. During most fish collections a backpack electrofisher or 2.5 GPP pram/boat electrofisher will be used. Because electrofishers send an electrical current through the water, not following safety procedures may result in serious injury or death. General safety guidelines include:

- Primary responsibility for safety while electroshocking rests with the team leader.
- **DO NOT CHASE FISH!!**
- All crewmembers should receive training in First Aid and CPR. Electro-fishing units have a high voltage output and may deliver dangerous electrical shock. Electric shock can cause heart fibrillations and/or death.
- While electrofishing, avoid contact with water unless sufficiently insulated against electric shock. Use chest waders with non-slip soles and watertight rubber gloves that cover to the elbow. If they become wet inside, stop fishing until thoroughly dry.
- Avoid contact with anode at all times. At no time while electrofishing should a crewmember reach into the water for any reason.
- The backpack electrofishing equipment provided is equipped with a 45-degree tilt switch, which interrupts the current. Do not make any modifications to the electrofishing unit, which would make it impossible to turn off the electricity.
- The pram/boat electrofishing equipment provided is equipped with an emergency shutoff switch. Do not make any modifications to the electrofishing unit, which would make it impossible to turn off the electricity.
- General safety guidelines should be observed. If waders or gloves develop leaks, leave the water immediately. Avoid operating electrofishing equipment near people, pets or livestock. Discontinue any activity in streams during thunderstorms or heavy rain. Rest if crew becomes fatigued.
- Gasoline is extremely volatile and flammable. Its vapors readily ignite on contact with heat, spark or flame. Never attempt to refill the generator while it is running. Always allow the generator to cool before refilling. Keep gasoline out of direct sunlight to reduce volatilization and vapor release. Always wear gloves and safety glasses when handling gasoline. Keep gasoline only in approved containers.
- Decision to use electrofishing equipment will depend on size of site, flow, conductivity and turbidity. If the specific conductivity is below 10 uS or > 1000µs; if the flow is too high; if the site is too deep; if the water is too turbid to assure safe footing or locate stunned fish, the crew may consider using the seine only or determine that site cannot be sampled. This is a safety decision.
- Formalin is a carcinogen and can also cause permanent damage to mucous membranes and eyes. Care must be taken when placing fish in formalin so that the fish does not flop around and splash formalin onto people near the jar. Proper precautions should be taken when handling formalin.
 - Protective gloves and eyewear should be worn
 - Avoid inhalation of vapors
- Always test water for depth and unseen structures.
- Never enter water that is flowing at an unsafe rate. Remember that slippery or loosely packed substrates may compound the effects of swiftly moving water. If you have any doubt about the safety of an area in the river, consult with the team leader. Never enter an area that is outside your personal comfort level.
- Never enter deep water alone.
- FAILURE TO OBSERVE SAFETY PROCEDURES WILL RESULT IN DISCIPLINARY ACTIONS INCLUDING PROBATION AND DISMISSAL.

4.0 Quality of the Measurement

4.1 Training

Principle investigators for the OWRB are required to have degrees and/or experience with biological or other applicable sciences. Principle investigators are defined as crew leaders, and this designation may be made upon the leader of a multi- or a one person crew. Training is required for all SOPs dealing with water quality and quantity collections and measurements as well as habitat assessments and biological collections. In-house training will be conducted for the use of all meters and digital titrators used for water quality or quantity measurements. Investigators must be familiar with OWRB SOP document and all training will follow the methods outlined in that document. Extra training will be provided when new SOPs are developed. Training of field crews will be done through dry run exercises in the laboratory to familiarize field crews with sample collection, sample preservation, instrument operation, calibration, and maintenance. In addition, when new personnel are hired or new methods developed, qualified staff will train on sample collection, measurement, and field analysis methods through side-by-side field trips. These trips will familiarize staff with SOP requirements. When training is considered adequate, a qualified staff member will check field staff for adherence to SOPs. Prior to collecting fish, all staff should familiarize themselves with Fisheries Techniques (edited by L.A. Nielsen and D.L. Johnson and published by the American Fisheries Society 1983), this SOP, and OWRB Technical Report 99-3 Standard Operating Procedures for Stream Assessments and Biological Collections Related to Biological Criteria and Development.

Investigators are tested for identification abilities with a statewide assemblage of fish fauna before fish collections begin. These fish are comprised of species that are typically found in Oklahoma stream systems. The majority of the test specimens include fish with larger body sizes that are typically field identified and/or found in large numbers. Species of special concern such as the Arkansas River Shiner are also utilized during the testing procedure to insure endangered or threatened species may be correctly identified and released. A test score on critical species of 95% or better must be achieved before the investigator will be a field crew leader. Investigators that score under 95% will not collect without direct supervision of the crew leader.

4.2 Kinds of Quality Assurance Samples

4.2.1 Replicate Collections

Replicate samples may be collected. The scope and number of replicates will be determined by the project Quality Assurance Project Plan. They may include replicates for various habitat or stream order.

5.0 Personnel and Equipment

5.1 Personnel

Fish collection crews will consist of two to three people. In some instances, a fourth crewmember may be added on larger streams where a pram or boat shocker is used. The team will consist of a team leader and one to three team members. The team

leader is someone with one or more seasons of collection experience who has scored above a 95% on critical species identification. Collection experience in other programs may be substituted for that with the OWRB. In certain instances, a team leader may have test scores below 95%. In this case, tests will be reviewed and species that were commonly missed in the scoring will be excluded from releasing. The team leader will have the final say on all crew activities. A team member is someone trained on fish sampling protocols. Team members will be expected to participate in the decision-making and follow the team leaders direction.

5.2 Equipment and Supplies

5.2.1 Backpack Electrofisher

A Smith-Root Backpack Electrofisher model 15 with a Honda model EX-350 generator will be used for collecting fish. Always use this equipment in accordance with manufacturer instructions. The team leader will provide a detailed explanation of how the shocker works as well as safety precautions. Each team member before operating and/or assisting with the shocker should read and understand the manuals for the generator and the shocker. Starting procedures, safety procedures and troubleshooting are well documented in these manuals and are not detailed here. The manuals can be obtained from the manufacturer. The shocking team must consist of at least two people—an operator and a netter. **This unit may be extremely dangerous if used inappropriately and without the proper safety equipment. Please refer to Section 2.0 for further details.**

The unit works to develop an electrical field, and everything within that field is subject to electric shock. The shocker consists of a trailing stainless steel cable electrode (cathode) and a ring electrode (anode) mounted on the end of a PVC pole. By pressing the anode button, the electrical field is created. The backpack electrofisher works effectively in specific conductance ranges from 40-1000 microsiemens (uS) with and optimal range of 150-650 uS.

The shocker selects for habitat and fish differently than does the seine. The electrofisher selects for fish with more surface area (i.e., larger or deep-bodied) such as bass or suckers. Also, the electrofisher is more effective in habitat where seining may be more difficult such as brush piles, root wads, undercut banks, bedrock ledges, cobble substrates, and shallow riffles. To effectively shock, the anode should be gradually passed back and forth over and in these areas as the team works upstream. As fish are stunned, they will usually roll over and become more visible, allowing the netters to see and capture them. Shocking may also be done in deep or shallow pools.

The unit should be maintained according to manufacturer specifications.

5.2.2 Pram or Boat Electrofisher

A pram or boat electrofisher may be used in waters with extremely low conductivity, conductivity greater than the backpack can handle (up to 1700 uS), and in deeper

waters. A Smith-Root 2.5 GPP electrofisher with a Honda model 2500 generator will be on the pram or boat. Always use this equipment in accordance with manufacturer instructions. The team leader will provide a detailed explanation of how the shocker works as well as safety precautions. Each team member before operating and/or assisting with the shocker should read and understand the manuals for the generator and the shocker. Starting procedures, safety procedures and troubleshooting are well documented in these manuals and are not detailed here. The manuals can be obtained from the manufacturer. The boat shocking team must consist of at least two people—an operator and a netter. The pram shocking team must consist of at least three people—an operator, a netter, and a pram guide. Because the guide must turn off the unit in case of accident, it is particularly important that this team member stay fully aware. **This unit may be extremely dangerous if used inappropriately and without the proper safety equipment. Please refer to Section 2.0 for further details.**

The unit works much like the backpack electrofisher, but certain distinctions do exist. The anode for the boat shocker is a set of arrays that are lowered into the water, and the anode for the pram is a wand but can be operated up to 100 feet from the shocking unit. The cathode for the both the pram and boat shockers is the either the boat if using an aluminum hull or a series of cathode arrays if using a fiberglass hull. When cathode arrays are used, they are mounted at the midpoint of starboard and port gunnels and the starboard and port corners of the bow.

The unit should be maintained according to manufacturer specifications.

5.2.3 Seine

Various sized seines may be used to collect fish. Recommended sizes vary from 3 to 6 foot seines in 10, 20, and 30-foot lengths. Seine height is dictated by water depth, and length is determined by width of the water being sampled. If possible the seine should be 15-25% longer than the width of the waterbody being sampled and about 25% higher than the depth of the water. This will allow the center of the net to form a bag behind the operators where the fish are more likely to stay in the net. However, it is important to remember that the longer the seine is, the more difficult it will be to control in stream currents. Therefore, rule of thumb for length may be discarded. When this occurs, extra time should be spent seining the missed habitats. In general, the OWRB uses 10X6 and 20X60. A seine 20X8 may be used in very deep and/or long pools. Seines should be a ¼ inch mesh to reduce fishing pressure on some young of the year.

The seine selects for habitat and fish differently than does the shocker. The seine selects for fish with less surface area (i.e., terete or smaller body plan) such as minnows or darters. Also, the seine is more effective in habitat where electrofishing may be more difficult pools or runs. Seining may also be done along banks and around instream cover. Seining technique is explained in the later methods section. Seining is often not the best method for collecting fish for toxics studies because of the selection for smaller fish.

Seines should be stored dry and free of debris and other snags.

5.2.4 Hoop Nets

Various sized hoop nets may be used to collect fish. Recommended sizes vary from 2 to 4 foot diameter nets with 4 to 6 hoops and ½ inch to 2-inch mesh. Hoop size is dictated by water depth, and fish being targeted determines length and mesh size. When in place, nets should be under the water and extended fully. To best target fish they should be placed near in-stream structure with the open end facing the structure being targeted. When placing the net, the cod end should always be secured with a zip tie and tied off so that fish cannot escape, and the open end facing downstream and tied to the structure being targeted. In general, the OWRB uses 2 and 3-foot diameter nets with 6 hoops and 1 to 2 inch mesh.

The hoop selects for habitat and fish differently than does the seine. It selects for fish with more surface area (i.e., larger or deep-bodied) such as bass or suckers. Also, the hoop net is more effective in habitat where seining may be more difficult such as deep water and heavy structure. For hoop nets to be most effective they should be used in association with instream structure and in deeper waters. They should also be in place for 12 to 24 hours and left overnight.

Nets should be stored dry and free of debris and other snags.

5.2.5 Gill Nets

5.2.6 Hook and Line

5.2.7 General Supplies

Clothing

Rubber Gloves	as many pairs as the shocking crew consists of
Waders	as many pairs as the shocking crew consists of, although everyone is responsible for their own waders
Goggles	for use in mixing formalin

Documentation

Camera with at least 2 rolls of film or adequate digital memory
Tape measure to record lengths of released fish

Chemicals

Gasoline/oil mix for generator
Extra two stroke oil

Shocker Parts

Spare plug, Plug wrench, and screwdriver
Spare Anode

Nets

- Fishing line or dental floss to repair nets
- Dip nets to collect shocked fish

Containers

- Foil for wrapping fish
- Large bags for placing fish in
- Adequate Ice Chests

6.0 Collection of Fish

The OWRB does not have the regulatory capacity to issue fish consumption advisories. Therefore this document is not meant to provide guidance for the collection of fish for this purpose. Fish collections for the purposes of toxic analysis as related to this SOP are screening studies and are strictly to determine the potential bioaccumulation of organics and heavy metals on certain fish populations in streams or rivers involved in special studies or of special concern. The analytical data is forwarded to the ODEQ for further processing.

6.1 Target Finfish Species

Toxics studies target a variety of finfish in Oklahoma (Tables 1 and 2). The species list was developed from various sources including Tables 3-1 and 3-3 of the EPA's Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories. It is important to note that not all species listed in Tables 1 and 2 may be considered sport fish or commonly consumed fish. Species are divided into two trophic categories—predator and bottom-feeders. Priority designates species to target in particular watersheds.

Table 1. Recommended Target Predator Species

Family Name	Common Name	Scientific Name	Priority
Lepisosteidae	Alligator Gar	<i>Lepisosteus spatula</i>	Low (uncommon)
	Spotted Gar	<i>Lepisosteus oculatus</i>	Medium
	Longnose Gar	<i>Lepisosteus osseus</i>	Medium
	Shortnose Gar	<i>Lepisosteus platostomus</i>	Medium
Salmonidae	Rainbow Trout	<i>Oncorhynchus mykiss</i>	High
	Brown Trout	<i>Salmo trutta</i>	Low (uncommon)
Esocidae	Northern Pike	<i>Esox lucius</i>	Low (uncommon)
Ictaluridae	Flathead Catfish	<i>Pylodictus olivaris</i>	High
Moronidae	White Bass	<i>Morone chrysops</i>	High
	Striped Bass	<i>Morone saxatilis</i>	High
Centrarchidae	Bluegill Sunfish	<i>Lepomis macrochirus</i>	Medium (small size)
	Smallmouth Bass	<i>Micropterus dolomieu</i>	High
	Spotted Bass	<i>Micropterus punctulatus</i>	High
	Largemouth Bass	<i>Micropterus salmoides</i>	High
	White Crappie	<i>Pomoxis annularis</i>	High
	Black Crappie	<i>Pomoxis nigromaculatus</i>	Medium
Percidae	Sauger	<i>Stizostedion canadense</i>	Medium
	Walleye	<i>Stizostedion vitreum</i>	High

Table 2. Recommended Target Bottom-Feeder Species

Family Name	Common Name	Scientific Name	Priority
Cyprinidae	Common Carp	<i>Cyprinus carpio</i>	High
Catostomidae	River Carpsucker	<i>Carpoides carpio</i>	High
	White Sucker	<i>Catostomus commersoni</i>	High
	Blue Sucker	<i>Cycleptus elongatus</i>	High
	Smallmouth Buffalo	<i>Ictiobus bubalus</i>	High
	Largemouth Buffalo	<i>Ictiobus cyprinellus</i>	High
	Black Buffalo	<i>Ictiobus niger</i>	Medium
	Spotted Sucker	<i>Minytrema melanops</i>	High
	River Redhorse	<i>Moxostoma carinatum</i>	High
	Black Redhorse	<i>Moxostoma duquesnei</i>	Low (small size)
	Golden Redhorse	<i>Moxostoma erythrurum</i>	High
	Shorthead Redhorse	<i>Moxostoma macrolepidotum</i>	High
Ictaluridae	Blue Catfish	<i>Ictalurus furcatus</i>	High
	Channel Catfish	<i>Ictalurus punctatus</i>	High

6.2 Composite Samples

Depending on available populations, a composite sample should be collected of one species from each trophic group. More than one species from each group may be collected if resources are available or if the study objectives warrant multiple collections. A composite sample will meet the following characteristics:

1. **Edible Size**—Generally fish should be at least 8-10 inches in length. If a size minimum or slot limit exists on a species for a particular area, the regulation should be considered when making collections.
2. **Comparable Size**—A composite should consist of fish near the same age. A good surrogate for age is length. Therefore, the smallest fish used in a composite sample should be no less than 75% of the total length of the largest individual.
3. **Collection Time**—Fish should be collected within one week of each other.
4. **Composite Size**—Composite should consist of 3 to 10 fish and be capable of yielding a homogenate sample of 200-g.

In some instances, more than one size range may be needed for each species. This will be specified in the study and be directed by the project supervisor. This is not a regular part of the collection methodology. In the instance that more than one size range needs to be collected, each size range will be treated as a different composite and will meet the characteristics described above.

6.3 Collection Methodology

The collection of fish follows may be done by any of the methods described in Section 5 of this document. Specific techniques for, and relative advantages for all methods vary considerably according to stream type and conductivity. The specifics are discussed in detail in Fisheries Techniques (edited by Murphy and Willis and published by the American Fisheries Society 1996).

The collection of fish involves the use of various collection methods including seining, electrofishing, hoop netting, gill netting, or hook and line. The combination of methods to be used at a particular site will vary according to site conditions. Variations of habitat, type of fish, and water chemistry dictate the use of different collection techniques. Electrofishing and hoop netting select for denser habitat (e.g., undercut banks, root wads, etc.) and very shallow riffles while seining selects for more open habitat such as runs or pools. Electrofishing and hoop netting select for more surface area (e.g., larger or deep-bodied fish) while seining selects for smaller fish such as minnows darters. Backpack electrofishers are not effective in waters with specific conductance <40uS and >1000uS. The 2.5 GPP electrofisher is not effective in waters with specific conductance over 1700 uS. Gill netting may be used in deeper waters without much structure. Hook and line fishing may be used in all habitats.

Sequence for fishing is determined by site characteristics. In general, shocking or hoop netting should be used first. These methods are selective for larger fish and may produce better results with less effort. Block nets may be used at all times. Seining should be done with minimal effort targeting certain habitats and not open waters unless a block net is set. Hook and line fishing should be a last resort. **RECORD THE TIME SPENT FOR EACH TYPE OF FISHING ON THE FIELD DATA SHEETS AS WELL AS NET SIZES.**

6.4 Sample Handling & Preservation

A composite will be considered a sample with each fish a part of the sample. The OWRB does not fillet or process fish in any other way in the field. All fish will be shipped whole to the analytical laboratory. The following steps should be taken to when handling the fish:

- **Before each use, ensure that all storage and measurement equipment has been scrubbed and rinsed to remove potential contaminants.**
- While fishing, fish should be kept alive until wrapped and placed on ice. The most efficient way to do this is to place fish of the same composite in separate filled tanks or fish bags.
- Care should be taken not to damage fish during collection or handling.
- Once fish have been collected, inspect and measure all fish and determine which will be included in each composite. Fish that have been heavily damaged by collection equipment should not be discarded and not included in the sample (this is the judgment of the team leader).

After collection has ended, fish need to be sorted, labeled, and preserved. Care should be taken not to cross-contaminate samples. Therefore it is important that each sample be processed separately, and that a different pair of plastic gloves be used for each sample. It may also be prudent to process a group of fish once the sample has been fully collected even if further fishing is necessary to collect the remaining sample(s). The following steps should be taken:

- **To avoid cross-contamination, each sample should be processed and handled consecutively with a different pair of plastic gloves being worn for each sample. Wash the measuring board between each sample.**
- **Sorting Procedure**
 - A positive identification should be made for all fish to be included in a sample. Fish of different species should be sorted into different groups.
 - Fish should be measured to determine that all fish lengths fall within 75% of each other. If a fish falls outside of the size range it should be released.
- **Labeling Procedure**
 - A label should be made for each fish in the sample and should include the following information:

Project ID _____	Date/Time _____
Collectors _____	County _____
Leg Des 1/4 _____ 1/4 _____ 1/4 _____ Sec _____ T.S. _____ R _____	
Station Name (ID) _____	
Coll. Met. _____ Fish # _____	
Trophic Level _____	
Common Name (Sp.) _____	
Length _____ Weight (optional) _____	
 - Record necessary information on the laboratory sample handling form.
- **Preservation Procedure**
 - Wrap each fish individually in foil twice ensuring that the fish is completely covered. Place the fish label between the first and second wrap.
 - Place fish in a double plastic bag. With an indelible marker, label the outside of the bag with the site name, date, and trophic group. More than one fish may be placed in the double bag. Remove all air from the bags.
 - Place fish on ice until they can be placed in a freezer. Fish should be fully covered in ice and remain frozen until processing.
- **Complete a Chain of Custody Form.**
- **Deliver to the analytical laboratory for processing.**

7.0 Forms

7.1 Field Notes

Field notes are documents used to annotate and record information that is gathered at the project site. They are a data sheet and should be treated as such. Therefore, they should be written, legible, and complete. To avoid confusion and loss of data, a new sheet should be used at each new project site. Field notes should be initialed and dated by the collecting personnel and data entry personnel. For guidance on proper procedure to complete the field notes, refer to your supervisor and or FTE. Field notes can be found at S:\Monitoring\STREAMS\forms\.

7.2 Laboratory Log-in Sheets

Log-in sheets are documents turned into the analytical laboratory for each sample collected. These forms are used to denote the parameters that should be analyzed. They are a data sheet and should be treated as such. Therefore, they should include the date and time of sample collection and be legible and complete. To avoid confusion and loss of data, a new sheet should be used at each new project site. For guidance on proper procedure to complete the log-in sheets, refer to your supervisor and or FTE. Log-in sheets can be found at S:\Monitoring\STREAMS\forms\.

7.3 Chains of Custody

Chains of custody are documents turned into the analytical laboratory for each group of samples collected. These forms are used for several purposes. They act as a legal document to show proper delivery of samples occurred and they make a general list of the parameters that should be analyzed. They are a data sheet and should be treated as such. Therefore, they should include the date and time for each sample collected and be legible and complete. They should also be signed and dated by field and laboratory receiving personnel at the time of delivery. To avoid confusion and loss of data, a new chain of custody should be used for each group of samples. For guidance on proper procedure to complete the chains of custody, refer to your supervisor and or FTE. Chains of custody can be found at S:\Monitoring\STREAMS\forms\.

8.0 Data Storage

All completed paper copies of forms and data sheets should be maintained with the appropriate station notebook. The data from the field notes and laboratory data sheets should be either entered into or uploaded to the Water Quality Biological Database. Each sample should be maintained electronically in the database under a unique sample number.

9.0 References

EPA (1995), Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories: Volume 1—Fish Sampling and Analysis, 2nd Edition, EPA 823-R-95-007, Office of Water, Washington, D.C.

Murphy, B.R. and D.W. Willis (editors) (1996), Fisheries Techniques. 2nd edition, American Fisheries Society. Bethesda, MD.

Oklahoma Department of Environmental Quality, Customer Services Division, (2003-2004), Personal Communication, Oklahoma City, OK.

Robison, H. W. and T. M. Buchanan (editors) (1992), Fishes of Arkansas, 5th edition, University of Arkansas Press, Fayetteville, AR.