

**SOP #EH-06**  
**Fish Collection by Seining or Electrofishing**

*(Adapted from Draft ERT/REAC SOP for Fish Collection)*

**TECHNICAL STANDARD OPERATING PROCEDURE**  
**FISH COLLECTION**

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**APPENDIX A: FISH HANDLING AND PROCESSING**

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### 1.0 SCOPE AND APPLICATION

Two methods which can be used to sample fish populations on hazardous waste sites are described below. These methods are applicable for fish collection in streams and shallow portions of lakes.

### 2.0 METHOD SUMMARY

A seine is an active netting technique that traps fish by encircling them with a wall of net. The bottom of the net is weighed down by lead weights or a leadline, and floats are attached to the top of the net. Many seines have a bag in the middle, where fish are concentrated as the net is closed.

Electrofishing uses electricity to capture fish. An electrical field is created in the water by passing a current between two submersed electrodes. Alternating current (AC) stuns fish in its field; they temporarily lose equilibrium and can be dip netted. Direct current (DC) will pull fish toward the anode, where they can be netted. Research objectives, habitat characteristics and availability of power source dictate the type of current to be used.

If fish are being collected for residue analysis, a minimum of 10 individual fish will be collected for each composite sample. If possible, similar species should be collected at each sampling area.

### 3.0 SAMPLE PRESERVATION, HANDLING AND STORAGE

If tissues are being analyzed for contaminants, fish should be kept on dry ice after processing. Fish for heavy metal analysis should be placed in plastic bags. Fish that are going to be analyzed for organic compounds should be wrapped in aluminum foil which has been rinsed with hexane and air-dried.

Fish collected for population studies can be preserved in ethanol or 10% formalin. Specimens should be stored in glass jars or buckets with non-rusting lids. Small fish can be fixed by simply placing them in ethanol or formalin. When preserving large fish, a slit should be made along the belly on the right side of the midline. Incisions should also be made in the dorsal muscle mass, on either side of the vertebral column. For proper fixation, the specimen volume should be no more than 50% of the total volume occupied by specimen and preservative.

Fish handling and processing activities will be conducted according to the procedures outlined in Appendix A.

### 4.0 INTERFERENCES AND POTENTIAL PROBLEMS

#### 4.0.1 Seining

Mesh size and length of a seine will determine size of fish which can be caught, and may affect how efficiently the seine can be pulled. Mesh sizes too small will be difficult to pull, especially if there is much debris in the water. High current velocity in a stream will also decrease seining effectiveness.

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To prevent fish from escaping under or over the net, it is imperative that the leadline be kept in contact with the bottom, and the float line must stay on or above the water surface. Streams or lakes with rocky bottoms or debris that snags the leadline will be difficult to seine effectively. Having a third person follow the seine and free it from snags helps prevent losing fish when the seine gets caught.

S seines can be torn as they are pulled through the water, leaving holes through which fish can escape. The seine should be inspected frequently, and repaired as necessary.

#### ***4.0.2 Electrofishing***

Environmental factors which can affect electrofishing include water conductivity, temperature, season, and time of day. Electrofishing success is poor in water with very high or low conductivity. Electrofishing is most effective in shallow habitats. If water temperatures are high, some fish species may move into deeper water where temperature is lower and oxygen is higher. During spawning season, some species may be captured in shallow areas that would normally be found in deeper areas. Electrofishing at night catches more species, larger individuals, and more fish than similar effort during the day.

Because batteries and generators used for electrofishing provide more than enough current to electrocute a person, it is vital that safety rules be observed. All members of an electrofishing crew should understand the system and the risks involved. One person should be in charge of the operation, and this person should control the power source. Shut down the power source before any repairs or equipment changes are made. Electrofishing should never be done alone, and the crew and power source should stay close together.

#### ***4.0.3 General***

Any time fish are collected, water and boat safety procedures must be followed. Wading can be dangerous, especially in swift currents or if the bottom is uneven or algae-covered. Samplers should always work in pairs, and wader belts should be worn to prevent waders filling with water if falls occur.

There is always a potential for drowning accidents when working around water. All field crews should include a person who is trained in CPR. When a person has stopped breathing, breathing must be restarted within 4 to 6 minutes. However, an attempt should be made to resuscitate anyone who has been submerged for up to one hour.

General guidelines for boating safety should be reviewed and followed for all activities which require transportation by boat.

When collecting fish on a hazardous waste site, field workers can be exposed to hazardous materials. Personal protective equipment should be worn to prevent exposure, and extra care should be taken to avoid falls into potentially contaminated water.

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**5.0 EQUIPMENT**

Equipment needed for fish collection is listed below, by procedure.

SEINING

Seine  
Buckets  
Carpet needle and string  
Waders  
Wader belts

ELECTROFISHING

Backpack electroshocker  
Battery  
Waders  
Buckets  
Wader belts  
Fiberglass handled dip nets

**6.0 REAGENTS**

No reagents are needed for fish collection if fish are being collected for residue analysis.

If fish are being collected for population studies, fish should be preserved in either 70% ethanol or 10% formalin.

**7.0. PROCEDURES**

**7.0.1 Seining**

Seine nets are constructed of mesh panels hung from a float line with a weighted lead line attached to the lower edge. Seines are selective sampling gear, and will not capture all sizes of fish. The size of fish you want to sample will determine the mesh size of the seine. Mesh size should be small relative to the target fish. Too large a mesh size will allow fish to escape through the net, however mesh sizes too small will be difficult to pull through the water. Seines are most effective in water no deeper than two-thirds the height of the net.

The net should have a pole at each end which is at least equal to the height of the net. Poles should be held at a 45° angle away from the direction of movement when pulling the seine.

For sampling a stream, the seine should be long enough to reach from bank to bank. Unless stream flow is very low, the seine is pulled against the current. Care should be taken to run the poles holding the seine directly along the bank, and under it if the bank is undercut. The leadline must remain in contact with the bottom to prevent fish from escaping under the net, and the float line must stay on or above the water surface. Several fish species (e.g. largemouth bass) will attempt to jump over the top of the seine when confined, so the float line should be above water when these are the target species.

After a collection is made, both seiners should walk onshore and pull the leadline up immediately. If there is no convenient place to beach the seine, the leadline can be lifted above water by both collectors at the same time. After the net is out of the water, captured fish should immediately be transferred to water-filled containers.

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In a lake, a seine may be pulled parallel to the shore or from offshore toward the shore. Alternatively, one end of the seine can be planted on the bank, and the other end can be pulled out, around, then back in to the bank.

### **7.0.2 *Electrofishing***

Use of electricity to capture fish is one of the least selective of all active fish capture methods. This method involves creating an electrical field in the water by passing a current between two submersed electrodes. There are two types of electrical current. DC always flows in one direction because the negative and positive ends (electrodes) of the circuit do not change. Direct current will induce galvanotaxis (forced swimming with orientation) and fish will move toward the anode. With ac, the anode (the positive electrode) and the cathode (the negative electrode) switch positions, so the current flows alternately in both directions. Fish exposed to ac will be stunned and lose equilibrium, and can be easily netted.

Electrofishing can be done using a backpack-mounted electroshocker unit, a shore-based unit, or from a boat. Backpack shockers are best for small streams. A minimum of three people are needed, one to run the shocker and two dip netters. The crew should wade upstream, with the dip netters beside or behind the electrode handler. All stunned fish, regardless of size or species, should be collected. The sampling area should be fished slowly and methodically, especially areas with in-stream cover. Captured fish should be placed in water-filled buckets. Nets can be set at the upper and lower ends of a stream section to prevent movement of fish out of the sample area.

Shore-based electrofishing is similar to backpack shocking, except that the power source stays onshore. Shore-based fishing is more dangerous, as voltages of shore-based units are higher than backpack units. The crew is also separated from the power source, and may not have safety switches. A buddy system should always be used during a shore-based electrofishing operation.

When electrofishing from a boat, the electrodes are suspended from a boom off the front of the boat. The boat should be driven slowly through shallow areas or along weed beds, and one or two people should stand near the bow and dip net stunned fish.

Research objectives, habitat characteristics and availability of the power source will influence the choice of current to be used. DC should be used when it is important not to damage or kill fish, and is very effective in turbid water or in thick weeds or brush. AC generators are generally less bulky, and are effective in clear unobstructed water. AC is more harmful to fish than DC, and may cause hemorrhaging, rupture swim bladders or fracture vertebrae.

Both direct and alternating currents can be modified to produce various current shapes that have different effects on fish. Pulsed DC will sustain forced swimming with less damage to fish. In addition, pulsed DC requires less voltage than ac and a smaller electrical source can be used. Pulsed ac will have the same effect as unmodified AC, but is not as potentially harmful to fish.

Water conductivity will affect the efficiency of electrofishing. In water where the conductivity ranges between 100 and 500 micromhos/cm, electrofishing will be most effective. At high conductivities, water

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is less resistive than fish and the current will flow around them. Electrofishing is not used in salt water habitats. Low conductivity water is more resistant than fish, and the electrical field is limited to the immediate area of the electrode.

### **8.0 CALCULATIONS**

No calculations are needed for the above procedures.

### **9.0 QUALITY ASSURANCE/QUALITY CONTROL (QA/QC)**

The following QA/QC procedures apply to fish collection and field processing:

1. All data will be documented on field data sheets (*see included example*) or in logbooks. Photo documentation will be done when possible.
2. Samples will be duplicated in an unimpacted reference area.
3. A sample plan specifying methods, target species, and sample size will be prepared before field work begins.
4. All deliverables will be peer-reviewed prior to release.

### **10.0 DATA VALIDATION**

Data generated will be reviewed according to the QA/QC considerations listed in Section 9.0.

If possible, species identifications will be confirmed by a regional biologist familiar with the site aquatic fauna.

### **11.0 HEALTH AND SAFETY**

All field crew members will conduct sampling in accordance with the appropriate level of health and safety training required by their parent organization.

Any time fish are collected, water and boat safety precautions must be taken. Wading can be hazardous in swift currents or if the bottom is uneven or algae-covered. Falls can be avoided by moving slowly, taking short steps, and wading sideways to the current. Guidelines for boating safety should be followed for all activities which require transportation by boat.

Safety procedures which should be observed while electrofishing include use of the buddy system, clear communication between the sampling team, and all samplers in waterproof gloves and waders which do not leak. The electrofishing equipment should be equipped with 'dead man' automatic shut-off switches, and one person should control the power source. At least one member of an electrofishing team must be certified in CPR.

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**APPENDIX A**

**Fish Handling & Processing**

*(Adapted from ERT/REAC SOP #2039 Revision 0.0)*

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**SCOPE AND DESCRIPTION**

This appendix describes the basic procedures for field processing of fish collected at hazardous waste sites. Fish can be used to determine whether contaminants in aquatic habitats accumulate in fish tissue, cause histopathological damage, or affect fish condition or growth. Impacts on aquatic community structure can also be assessed.

**METHOD SUMMARY**

Specific procedures used to process fish will depend on the project objectives. Regardless of the objectives, data which should always be collected on fish in the field include length, weight, species, and information on parasites or other abnormalities. When possible, sex and stage of maturity should also be noted.

Fish which are collected for contaminant analysis should be measured, then filleted or frozen whole. If study objectives include histopathology, fish should be dissected so sections of target tissues can be collected.

**INTERFERENCES AND POTENTIAL PROBLEMS**

**General**

Extreme temperatures can alter tissue characteristics, making tissues unsuitable for analysis. Exposure of dead specimens to extreme cold can cause tissue to freeze, making histopathological analysis difficult. Extreme heat can cause rapid decomposition of tissue. An effort should be made to keep fish alive until they are processed. Dead fish should be processed as soon as possible.

All members of the processing staff should be trained in techniques used to make length and weight measurements. Inconsistencies in the way these measurements are taken can lead to errors.

In some cases, fish collected may not have sufficient body mass for analysis of a contaminant to a given detection limit. If this occurs, then individuals of the same species from the same sampling location may be pooled for analysis. If multiple analyses of contaminants in tissues are being done, these may need to be prioritized if body mass of the specimens is insufficient to conduct all of the analysis. Analyses to be conducted on each specimen should be carefully documented.

**Length**

Factors which contribute to length measurement errors are muscular tension in live fish, eroded fins, shrinkage of fish due to preservation, and failure to consistently squeeze the tail to get maximum total length.

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**Weight**

When taking weights, an attempt should be made to have fish at a standard degree of wetness. Variation in stomach contents or amount of water swallowed at capture will also affect fish weights. Other sources of error include movement of the scale due to fish movements, wind or boat motion.

**EQUIPMENT/APPARATUS**

Equipment needed for processing fish is listed below:

Data Sheets	Measuring board
Balance or scale	Field guides or keys
Coin envelopes	Knife
Forceps	Saw
Probe	Pliers
Ziploc® bags	Aluminum foil
Large scissors	Small scissors
Dissecting microscope	Glass scintillation vials with lids
Glass jars with lids	Preservative
Scalpel	Fillet knives
Knife sharpener	Dissecting trays

**REAGENTS**

No reagents are needed for fish processing if fish are being collected for residue analysis. Tissue sections collected for histopathological analysis should be preserved in glass scintillation vials filled with 4 percent buffered paraformaldehyde. Buffered paraformaldehyde can be purchased through commercial chemical supply companies. Tissue sections for histopathology should be collected before fish are frozen. Fish being collected for population studies can be preserved in either 70 percent ethanol or 10 percent formalin.

**PROCEDURES**

When fish are collected for residue analysis, generally the largest fish captured are the ones which should be analyzed. All animals captured should be held either until a sufficient number and weight of fish are caught at a station, or until the end of the day. If necessary, fish should be marked or tagged as they are captured so that individual fish can be identified later. Length, weight and species should be determined at the time a fish is tagged. Other data can be collected after fish which will be analyzed have been selected.

A data sheet should be completed for each specimen processed. Sampling location, tag number, date, species, and data on the specimen metrics described below should be recorded.

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**Length**

Fish length is measured using a measuring board on which the anterior end of a fish is placed against a stop at the beginning of a measuring scale. The fish should be measured with mouth closed, and the body positioned on its right side with the head to the measurer's left. Any one of three measurements can be taken: total, fork or standard length (Figure 1). Total length is the greatest length of a fish from its anterior most extremity (usually the mouth) to the end of the tail fin. For fish with a forked tail, the two lobes should be pressed together, and the length of the longest lobe should be taken. Fork length is measured from the anterior end of the fish to the tip of the middle rays of the tail. Standard length is the length of a fish from the anterior end of the fish to the tip of the middle rays of the tail. Standard length is the length of a fish from the anterior end to where the base of the median tail fin rays join the caudal peduncle. This spot can be located by bending the tail sharply. A crease should form where the tail fin rays end. Total length or fork length measurements are used most often. Determination of standard length is very difficult on some species.

**Weight**

Spring balances or electronic digital scales are generally used to weigh individual fish. Fish can be weighed by themselves, or by placing them in a container of water. Taking the weight in water reduces error due to fish movement, but may not be practicable for large fish. Large numbers of fish can be weighed in bulk if individual weights are not needed (e.g., for population studies).

Because most fish maintain near-neutral buoyancy in water, their specific gravity is close to 1.0 and body volume is proportional to weight. Therefore, the amount of water displaced in a container can also be used to determine weight.

**Species Identification**

Study objectives will dictate what level of identification is needed for a fish. Fish collected for residue analysis should be identified to species, as different genera may have different feeding habits.

Local authorities should be consulted before field work begins to determine whether regional taxonomic references exist.

**External Examination**

While processing fish, note any external abnormalities or parasites on data sheets or in field logbooks. Information on sex and stage of maturity should also be noted. If fish are collected during spawning season, some fish can be sexed based on breeding colors. Mature fish may release eggs or milt when they are handled.

**Final Processing**

To assess environmental risk through food chain concentration of contaminants, the whole body should be analysed for tissue residue. Based on the objectives of the study, the stomach contents of the fish may

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be removed (using dissection technique) prior to analysis. Alternately, fish may be held in aerated chambers for 24 hours to deplete stomach contents. This will allow for a determination of the concentration of contaminants accumulated in the tissue versus contaminants entrained in the gut.

To assess risk to humans from fish consumption, the fish should be filleted and only muscle tissue sent to the laboratory for analysis. Fish should be dissected if tissues are being collected for histopathology or for residue analysis on specific organs.

Procedures for filleting or dissecting a fish are described below. Fish should be killed by a blow to the head immediately before processing.

Filleting

To fillet a fish, an initial cut should be made from the dorsal fin to the pelvic fin, just behind the opercular flap (Figure 2). Run the tip of the knife along the dorsal side of the fish, from the initial cut to the caudal fin. Continue making successively deeper cuts, running the knife blade as close to the neural spines and ribs as possible. After the fillet is obtained, remove the skin. Place the skin side of the fillet down on the dissecting tray, hold on to the tail portion of the fillet, and run the knife between the skin and the muscle tissue. Turn the fish over and repeat the process to obtain the other fillet.

Dissecting

Begin the dissection by laying the fish on its right side and making an incision from just above the vent to the top of the rib cage. Cut along the rib cage, forward through the pectoral girdle. Make a shallow incision to avoid damage to internal organs. Pull the flap downward to open the body cavity. Note any gross abnormalities or parasites observed in the body cavity. Also record sex and stage of maturity.

Liver, gill and kidney tissues are the fish tissues collected most often for histopathology or residue analysis. The liver should be located near the anterior end of the stomach. It is connected to the gut by the gall bladder and bile duct. The liver should be removed and weighed to the nearest 0.001 g. A hepatosomatic index, liver weight expressed as a percentage of body weight, can be used as an indicator of fish condition. For histopathology, two tissue sections should be obtained from the distal end of the medial lobe. The sections should be cut 1.0 centimeter (cm) towards the center of the lobe, and 0.5 cm thick. Cut the section using a scalpel, and handle carefully to avoid crushing the tissue. Place the tissue sections in a glass scintillation vial filled with 4 percent buffered paraformaldehyde.

The gills are located beneath the opercular flap. Pull back or remove the operculum to expose the gills. Carefully remove a section of gill tissue, taking care not to crush it. Place the gill tissue in the scintillation vial with the liver tissue.

The kidney is located along the backbone above the gas bladder. Kidney tissue is difficult to remove from fish because it adheres to the body wall and is soft. Thin slices can be taken through the vertebral column which include the kidney. These tissue sections should be preserved with the liver and gill tissue sections. Again, for proper preservation, the specimen volume should be no more than 50 percent of the total volume occupied by specimen and preservative.

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Unless specific organs are being analyzed for residues, place all tissues back in the body cavity and wrap the fish in plastic or aluminum foil. Samples should be labeled and shipped following procedures outlined in the Sample Documentation and Sample Packaging and Shipping SOPs.

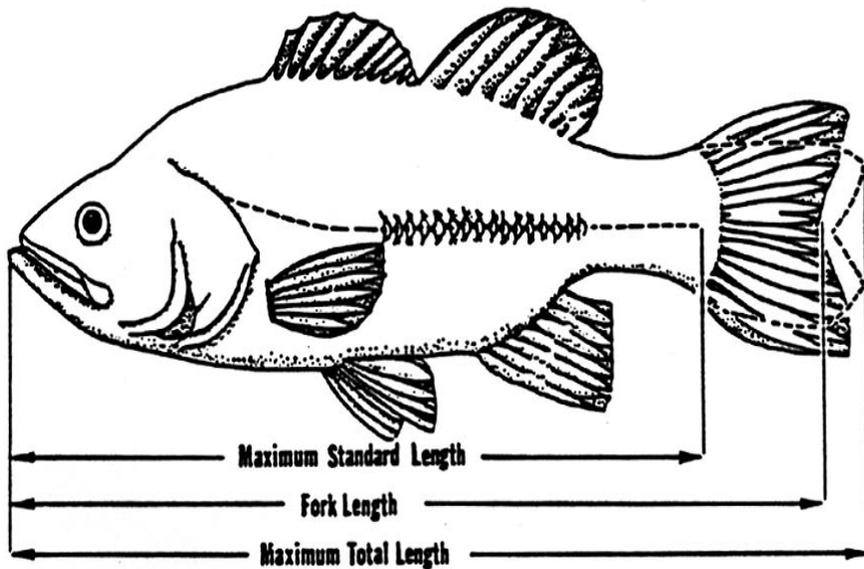
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FIGURE 1. Measurements of Fish Length - Standard, Fork, and Total  
(From Anderson and Gutreuter 1983)



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FIGURE 2. Location of Cuts for Filleting a Fish

