Protocols for Sampling Fish Communities in Wadeable Streams and Rivers

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Standard Operating Procedure
Methods for Sampling Fish in Wadeable Streams and Rivers

1. **Applicability.** This standard operating procedure (SOP) is used by the Division of Environmental Assessment for sampling fish in wadeable streams with either seine nets or portable electrofishing equipment. This procedure will provide for efficiency in sampling effort while maintaining safety of the technicians and fish.

2. **Purpose.** The method of sampling fish with a seine provides "screening level" information about the presence of fish species in a stream. In contrast, the electrofishing method provides information about the abundance, length, and weight of fish species in a stream.

3. **Definitions**
   A. **amp:** an abbreviation for ampere, which is the unit of measure expressing the quantity of electricity flowing through a circuit
   B. **anode pole:** An electrode pole with the switch that activates the electrical current and functions as the + terminal (the anode attracts anions and electrons)
   C. **"bag":** A description of the shape of a seine when moving water makes the net swell or "inflate"
   D. **cathode pole:** An electrode pole without the switch that activates the electrical current and functions as the – terminal (the cathode attracts cations and positive charges)
   E. **conductivity:** A measure of the ease with which electricity can flow through water (units are micro-Siemens per centimeter, µS/cm)
   F. **current:** a generic term that refers to the flow of electricity through a circuit (unit of measure is an amp). The convention is that the direction of the current is the flow of positive charges from the + terminal to the – terminal. The flow of electrons is the reverse direction from the – terminal to the + terminal.
   G. **DC:** direct current electricity
   H. **duty cycle:** The percent of time that the electrical circuit is producing an electrical current. For example, if the duty cycle is 20% then the circuit is on for 0.20 seconds and off for 0.80 seconds.
   I. **electrofisher:** The portable backpack electrofishing unit
   J. **frequency:** The number of electrical pulses per second (unit of measure is a Hertz (Hz)). For example, a 30 Hz frequency would produce 30 pulses per second.
   K. **narcosis:** State of electrically-induced immobility with slack muscles, which is desirable while electrofishing
   L. **pulsed DC (PDC):** pulses of direct current electricity
   M. **"rat tail":** A cathode consisting of an uninsulated strand of metal cable

N. **seine:** A rectangular net used to collect fish. A seine typically attaches to poles on both sides, has a series of weights on the bottom, and a series of floats on the top.
O. **specific conductance:** a standardized measurement of conductivity where the conductivity is adjusted to a standard temperature of 25°C.
P. **shocker:** The person that operates the electrofisher
Q. **taxis:** (aka electrotaxis) is electrically induced movement, usually toward the anode, which is desirable while electrofishing
R. **tetany:** state of electrically-induced immobility with rigid muscles, which is not desirable while electrofishing
S. **volt:** The unit of measure of the amount of "pressure" in an electrical current. This is analogous to the pressure of water in a flowing through a hose.
T. **"wand":** A synonymous term for anode pole
U. **watt:** the unit of measure for electrical power, with the abbreviation W (1 Watt = 1 Amp x 1 Volt)

4. Responsibilities

A. **Training**. It is the responsibility of the team leader to ensure that the individuals participating in the sampling are trained in the proper and safe use of the equipment. For electrofishing, team leaders must complete an intensive training course on the use of electrofishing gear, such as the *Principle and Techniques of Electrofishing (Online) - CSP2C01* offered by the U.S. Fish and Widlife Service (https://nctc.fws.gov/courses/CSP/CSP2C01/resources/).

B. **Data recording**. It is the responsibility of the individual collecting the data to record the results and additional qualifying information on standard field sheets obtained from the MDEP Biomonitoring program.

C. **Data submission**. It is the responsibility of the team leader or the staff member collecting the data, as appropriate, to place completed field sheets in the appropriate field sheet folder located in the Biomonitoring staff area.

5. Guidelines and Procedures

A. **Sampling period**. Fish sampling typically occurs from June to September. Fish sampling in the spring can be complicated by high water levels. Fish sampling in the autumn can be complicated by leaves fallen from trees that obscure fish.

B. **Notifying other State agencies**. **Sampling fish must only occur as part of a Scientific Fish Collectors (SFC) permit administered by Maine IF&W**. Anyone handling the fish, or the equipment must be on the SFC permit. Sampling is only allowed in waterbodies that are identified in the permit. The team leader must ensure that all conditions of the SFC are followed, including written notification of Regional Biologists

and District Wardens prior to sampling when required. This may be accomplished by emailing the secretary at the regional office. Make sure to receive an acknowledgement of the notice prior to sampling, and if none is received, make direct contact with the biologist and warden DMR salmon biologists must be consulted in advance for streams that may have salmon before any sampling occurs

C. **Fish health.** The primary goal is to collect valuable data while minimizing any stress to the fish. The fish should be handled with care and respect. Staff should minimize the amount of time that the fish are out of the water. Monitor any fish in buckets or tubs and if signs of distress are evident, process or release immediately, change the water, or take other measures to reduce stress. Never handle trout with a dry hand, which can result in loss of protective mucus and lead to disease.

D. **Seine method**.

(1) Equipment and personnel. This method requires a 5' to 8' wide minnow seine with poles on either end. This method requires at least 2 people but it can be helpful to have a third person. The seine consists of rectangular net with a weighted bottom edge and floating top edge and usually poles on each end.

(2) Applicable habitat. This method is appropriate in shallow water, typically less than 75 cm deep, where the morphology of the channel concentrates fish and minimizes escape routes for the fish. The method works best when sampling toward a channel feature that blocks fish movement and forces fish into the seine. This method does not work well in 1) wide streams where the fish can simply swim away from seine, 2) deep streams, 3) streams with a lot of boulders.

(3) **Using the seine**.

(a) A person holds the pole at one end of the net and another person holds the other pole. While standing next to the pole, the hand that is on the same side as the pole grips the top part of the pole and the other hand grips the middle of the pole. The edge that floats should be on top so they will float on the surface of the water.

(b) The people at either end of the net should be close enough that the net forms a U-shaped bag with the net "inflated" by either the natural, downstream movement of water or by the people dragging the net through the water. In addition, the poles should be held so the pole is angled with the top of the net behind the bottom of the net to facilitate the formation of the "bag" (approximately 45° angle). The bottom of the poles should be on or near the bottom of the stream at all times to minimize gaps under the net. The U-shaped bag is important for two reasons. First, it allows the bottom of the net to rest on the bottom of the stream and better conform to the contours of the substrate. If the net is stretched taught, then gaps will appear under the net, providing escape routes for fish. Second, the U-shaped

bag allows an area for fish to swim into the net and subsequently get captured. An ineffective bag can be caused by having the poles too far apart, thereby stretching the net, or by having the poles too upright (perpendicular to the stream bottom).

(c) Seining works best when sampling from downstream to upstream so the water current "inflates" the net to form what is called "the bag". In some cases, it is advantageous to sample perpendicular to stream flow to trap fish along a bank or tree roots. Sampling from upstream to downstream in the same direction of the water flow generally does not work well if the water velocity if fast enough to "deflate the bag". In other words, if the downstream flow of water is faster than the people walking downstream then the bag may collapse and lose its shape.

(4) **Sampling strategies**. The probability of successfully using any of these methods is increased by making sure you form a good "bag" and minimizing gaps underneath the net.

(a) Sampling a pool. Start at the downstream part of the pool and sample in an upstream direction while attempting to trap fish at the upstream end of the pool. If available, additional people can walk alongside the net to herd fish and prevent them from swimming around the net. At the upstream end of the pool, the additional person could stomp around to scare fish into the net.

(b) Trapping fish along a bank. This strategy targets habitat provided by an undercut bank or tree roots. Start opposite the bank and walk across the stream to the other bank. Trap the fish in the target habitat being careful to minimize escape routes. Have a third person agitate the water in the target habitat and attempt to scare fish into the net. Alternatively, use one pole to jab around a little and scare fish into the net. Be ready to raise the net quickly. This method has the greatest potential of scaring a large trout or sucker into the net and they quickly try to escape the net.

(c) Sampling a riffle. Hold the net in a riffle area and make sure to have a good bag. Have one of the people holding the net or a third person kick around the area in front of the net. This approach targets species that particularly like riffles, such as sculpins, long-nose dace, and salmon parr.

(5) **Catching fish**.

(a) When fish are in the net, the people holding the poles should communicate and pull the net up at the same time. The bottom hand does most of the work by lifting the pole in forward and upward motion. It helps to step forward during this motion to maintain balance and leverage. When the bottom edge of the net nears

the surface of the water, then use both hands to lift the poles straight up. Allow a portion of the net to remain submerged so the fish are still in the water.

(b) The person responsible for identifying the fish should move their hands from the pole to grasp the net. Slowly alternate releasing and grabbing further up the net toward the other person to decrease the amount of net between the two people. Be aware of any fish that might be out of the water and attempt to keep fish in the water.

(c) When the area of net between the two people is sufficiently small, then the person responsible for identifying the fish can reach into the net and carefully grab one fish at a time. Since this is just a screening tool focused on species occurrence, do not worry about counting the fish. Rather, focus on identifying all the species that are encountered. Return the fish to the water after identifying them.

(d) Record the species that are present on the field sheet. If salmonids are present, then indicate the age classes present.

(6) **Cleaning the seine**.

(a) After sampling, hold the net out of the water and attempt to remove large debris from the net.

(b) Hold the poles horizontally and stretch the net. Vigorously move the poles up and down to shake the net up and down. Rotate the poles and net and repeat the process on the other side of the net.

(c) Hold the poles vertically with the net completely out of the water. Have one person walk toward the other person. When approaching the other person, the person that is walking should grab the net near the closest float and drape it over the top of the pole. Do this for each of the floats. When the person finishes this process, then hold both poles horizontally and roll the net up.

(d) After returning to the truck, follow the Division's decontamination procedure.

(e) After returning to the boat barn, check to see if the net is damp and unroll it as necessary to help it dry out.

E. **Electrofishing method**

(1) **Equipment**

(a) Backpack electrofishing unit, 2 fully charged batteries, and electrode poles

(b) Waders or hip boots. Staff must wear waders or hip boots made from rubber or similar material. Staff must not wear equipment made from "breathable" materials. Waders or hip boots with felt bottoms are preferred for better traction. Avoid waders with metal studs in the soles.

(c) Three to four pairs of rubber "electrician" gloves. All people must wear the rubber gloves when the electrofishing unit is turned on.

(d) Nets with long fiberglass or wood handles

(e) Two or three 5-gallon buckets

(f) Portable battery-powered scale for measuring fish weight

(g) Four or more small, buckets

(h) Measuring board

(i) Credit card or similar shaped piece of plastic to help pick up small fish from measuring board

(j) Field sheets, clipboard, and pencils

(k) GPS unit

(l) Camera

(m) Vials, jars, and formalin in case it is necessary to collect a voucher specimen

(n) Water quality meters (e.g., pH, specific conductance)

(o) Cushions for sitting on while measuring the fish (optional)

(p) Extra batteries

(q) Polarized sun glasses

(2) **Personnel**. The team should consist of 3 or 4 people

(a) Shocker: the person that operates the electrofisher

(b) Primary netter: the person responsible for catching the most fish

(c) Secondary netter: this person is responsible for backing up the primary netter, carrying the bucket of fish, and keeping track of distance from the downstream

marked point. The secondary netter is typically the least experienced member of the crew. The secondary netter is also responsible for observing the fish in the bucket and alerting the operator if the fish show signs of stress, such as "branding" marks or remaining stunned for too long.

(d) Bucket person (optional): if a fourth person is available, then that person can assume responsibility over carrying the bucket of fish and keeping track of distance from the downstream marked point. This arrangement will allow the secondary netter to focus just on catching fish.

(3) **Preparing the electrofisher**. Follow the directions in the manual specific to the electrofisher being used, currently a LR-24 Electrofisher.

(a) Remove the protective cover on the electrofisher

(b) Install the battery

(c) Attach the anode pole, which is the electrode pole with the switch

(d) Attach a cathode, which can either be cathode pole or rat tail. The rat tail is preferred because of its greater surface area. It is safer for the fish if the surface area of the cathode is greater than the surface area of the anode. One disadvantage of the rat tail is that the tip of the cable can produce an intense electrical field.

(e) Replace the protective cover

(f) Have the person operating the electrofisher put it on their back (adjust straps as necessary)

(4) **Activate and calibrate the electrofisher** (typically the primary netter's responsibility)

(a) Make sure that everyone is wearing protective boots and gloves and tell them that you are turning the electrofisher on

(b) Pull up on the red safety switch on the top of the electrofisher

(c) Reset the timer

(d) Instruct the operator to place the anode and cathode in ankle deep water approximately 12" apart

(e) Run the calibration procedure and adjust (Table 1)

- Measure conductivity. Measure the conductivity of the water with the water quality meter. If the water quality meter measures both ambient conductivity and specific conductance, then focus on the ambient conductivity at this point. Specific conductance standardizes conductivity measurement to a standard temperature of 25°C. The actual ambient conductivity is what we care about for electrofishing.

- Voltage. There is an inverse relationship between the specific conductance of water and the amount of voltage necessary to momentarily stun fish. It is easier to stun fish in water with higher conductivity because the electrical current has an easier time travelling through the water. Electrical currents have more difficulty travelling through water with lower specific conductance, thus a higher voltage is needed to stun the fish. The automatic calibration procedure tends to underestimate the voltage needed to effectively stun fish without hurting them. Start by increasing the voltage by approximately 10% of the recommended voltage. The voltage can be incrementally increased **Higher voltages can increase the risk of fish injury. Avoid setting the voltage greater than 500 V. In general, lower voltages are safer for fish than higher voltages.**

- Frequency. Set the frequency to 30-60 Hz. Use 30 Hz in streams without salmonids. Use 60 Hz in streams with mostly salmonids. Use 50 Hz for streams with a mixed community of fish that include salmonids. **Higher frequencies can increase the risk of fish injury. In general, lower frequencies are safer for fish than higher frequencies.**

- Duty cycle. Set the duty cycle to 12% to start. The duty cycle can be incrementally increased to a maximum of 50% if fish are not responding to the electrical field.

- Wave form. Direct current is preferred in streams with conductivity <115 µS/cm. Pulsed current can be more effective in streams with higher conductivity. Direct current is thought to be safer for the fish.

Table 1: Selecting the proper settings for the electrofisher

Setup procedure	• Measure the ambient conductivity and consider DC waveform if conductivity is <100 µS/cm • Run automatic setup procedure and bump of voltage by approximately 10% • For salmonids, first try 12% duty cycle and 30 Hz frequency and bump up to 60 Hz frequency if necessary. PDC waveform may work better with adult salmonids because of it inducing taxis (movement toward the anode). • For bass and sunfish, try 20-30% duty cycle and 60 Hz frequency • Incrementally increase voltage as necessary but do not exceed 500 V • If there is insufficient response from fish as 500 V, then bump the voltage back to the original setting, increase the duty cycle, and then incrementally increase the voltage again
Signs that the fish are not being shocked enough	• avoidance • escape swimming
Signs that the fish are being shocked the correct amount	• taxis (fish are swimming toward the anode, often in a curve) • narcosis (fish are immobilized but have relaxed muscles) • gentle curvature of the body
Signs that the fish have been shocked too much	• false taxis (fish is unconscious with twitching muscles, often belly up, "false swimming") • tetany (fish are immobilized but have rigid muscles) • strongly bent body • flared gills • discoloration and branding • take more than 15 seconds for fish to recover
General rules of thumb	• a common assumption is that the conductivity of fish is around 115 µS/cm • in theory, larger fish are more susceptible to electrical fields than smaller fish because they experience a greater head-to-tail voltage gradient • the strength of the electrical field is greatest next to the electrodes and dissipate • the greatest risk for fish tetany is near the electrodes • duration of exposure to electrical field increases risk of injury • DC is safer for fish than PDC • DC drains battery power much faster than PDC • as duty cycle is decreased, increasingly higher power densities are needed to immobilize fish and there is a greater risk of tetany

	• the primary PDC attribute leading to injury is high frequency settings
Netting strategy	• net large fish and fish closest to the electrodes first • try to net fish before they get close to the electrodes • net fish as quickly as possible • avoid reintroducing captured fish back into the electrical field

(5) **Sample fish with the electrofisher** (Table 2)

(a) The secondary netter should capture the coordinates of the downstream end of the sample reach with the GPS unit, change the mode on the GPS unit to display the distance from the downstream point, and fill the bucket with 3-4" of stream water

(b) Walk upstream and activate the electrofisher in habitat that might be good for fish, such as pools, riffles, undercut banks, and submerged tree roots

(c) The anode and cathode should be underwater whenever the unit is activated. The anode is often placed upstream of the cathode. The person carrying the unit should hold the anode pole 2' to 6' away from the cathode. Avoid moving the anode and cathode too close together while activated because the intense electrical field resulting from that arrangement can harm fish. Fish may be attracted to the electrodes, particularly the anode, when activated. Thus, the anode can be used to coax fish from undercut banks, vegetation, and other difficult habitat.

(d) The primary and secondary netters typically flank the shocker with the secondary netter a little further downstream. If the current is fast, then both netters may stand downstream of the shocker. The primary netter may stand upstream of the shocker when the shocker is at the downstream edge of a pool.

(e) In streams segments with gentle water velocity, the netters must actively try to scoop fish with the nets. In streams with stronger water current, the nets may be place downstream, perpendicular to the flow so fish may float into the nets.

(f) Transfer captured fish from the nets to the bucket of water

(g) Stop sampling when you are 50 m from the downstream point and mark the upstream point with the GPS

(h) Record the wand time, voltage, frequency, wave form, and duty cycle on the field sheet

Table 2. Troubleshooting Guide for Electrofishing

Problem	Cause	Potential Remedy
Fish are swimming away from the electrical field and are not getting stunned	Electrical field is not strong enough to attract and immobilize fish	• Increase voltage (a little at a time) • Switch to pulse DC waveform • Increase frequency up to a maximum of 60 Hz
Fish are remaining stunned for over a minute in the bucket or show signs of injury (e.g., patches of discoloration)	The fish were shocked too much	• Decrease voltage • Switch to continuous DC waveform • Decrease frequency down to a minimum of 30 Hz • Reduce the amount of time fish are in the electrical field • Avoid touching the fish with either the anode or cathode
Fish are dead or have discolored patches ("burn marks")	The fish were shocked way too much	• Decrease voltage a lot • Switch to continuous DC waveform • Reduce the amount of time fish are in the electrical field • Avoid touching the fish with either the anode or cathode
The beeping sound emitted by the electrofisher is too quiet	The black disk on the side of the electrofisher near the top of the unit is oriented so it is muffling the sound	• Rotate the black disk to reveal the holes that let the beeping noise out
Stunned fish are stuck in crevices between rocks	Bad luck	• **DO NOT reach into the water with your hand** • **DO NOT attempt to remove the fish with a powered electrode pole because it could harm the fish** • Try to gently dislodge the fish by 1) swirling the water near the fish with a net or 2) touching the fish with a net or unpowered electrode pole

(6) **Weigh and measure the fish**

(a) Set up the scale and measuring board where there is a comfortable place to sit

(b) Make sure that the fish have sufficient fresh water and change water as necessary

(c) The first person is responsible for identifying the fish and measuring fish lengths and weights

(d) The second person is responsible for recording fish lengths and weights on the field sheet

(e) Depending on the crew and number of fish, the third person may help in the following ways:

- Sort fish by removing them from the large bucket and place them into smaller buckets, grouped by species
- Collect water samples (typically alkalinity)
- Operate water quality meters
- Capture digital images with the camera
- If there are a lot of fish, then this person may take over the responsibility of weighing the fish

(f) Priority for processing fish

- salmonids
- fish that are sensitive to handling, such as sculpins, common shiners, and northern redbelly dace
- fish that are more tolerant of handling, such as creek chubs, blacknose dace, white suckers, and sunfish

(g) The first person removes fish from a bucket, identifies them, and then measures their length with the measuring board. Make sure that that the measuring board is moist. Place the fish's snout against the side of the measuring board at 0 cm. Measure the total length in centimeters from the tip of the nose to the tip of the tail fin. Use a plastic card to help remove small fish from the measuring board. Fish less than 25 mm in length are not included in the tabulation, except for small shiners such as bridle and blacknose shiners.

(h) Weigh the fish in a yellow bucket on the scale. Fill the yellow bucket with a little bit of water and place it on the scale. Hit the Zero button to set the weight of the bucket of water to 0 g. Place the fish in the bucket, allow the weight to stabilize, and record the weight in grams. Shield the bucket and scale from the wind as much as possible so the wind does not make the weight fluctuate. If the wind is a

factor, then try filling the yellow bucket with a lot of water. Make sure to zero the weight between fish measurements or after dumping fish and getting fresh water.

(7) **Packing up gear at the truck**

(a) Follow the Division's decontamination procedure by inspecting nets, buckets, and boots, removing debris, and applying disinfectant spray as necessary

(b) Detach the anode and cathode cables and remove the battery

(8) **Caring for the gear at the end of the day**

(a) **Always recharge the battery at the end of each day of use (the battery will lose its capacity to retain a full charge over time if it is not charged after every use)**

(b) Spread out or hang gloves and boots so they will dry