APPENDIX A

STANDARD OPERATING PROCEDURES

FOR THE

BOAT ELECTROFISHING POPULATION SURVEY



This document describes the procedures for ORSANCO's Boat Electrofishing Population Survey and provides guidelines for the proper collection and processing of Ohio River Basin Fish harvested during electrofishing activities. This SOP has been developed to maintain continuity and ensure data quality.

1.0 Field Equipment

The following equipment is used to conduct electrofishing surveys:

1.1 Electrofishing Watercraft

- 1.1.1 19-ft. aluminum johnboat w/ 115hp and 25-30hp outboard motors
- 1.1.2 MLES® Infinity Electrofishing System w/ 5000-7500W gasoline generator
- 1.1.3 Dual, Retractable Anode Array
- 1.1.4 Minimum (4) LED DC flood and spot lighting array
- 1.1.5 Positive pressure cut-off foot pedal switch
- 1.1.6 50 75-gallon plastic or aluminum live well with aerator or flow-through system
- 1.1.7 Marine radio
- 1.1.8 Personal LED headlamps
- 1.1.9 12-Volt 2+ million candlepower spotlight

1.2 Fish Collection

- 1.2.1 Fiberglass handled nets with 0.25-in mesh
- 1.2.2 Rubber-soled footwear for each crew member
- 1.2.3 Personal Floatation Device for each crew member
- 1.2.4 Gloves for each crew member
- 1.2.5 Ear and eye protection for each crew member

1.3 Fish Processing

- 1.3.1 Fish identification reference keys
- 1.3.2 Fish measuring board
- 1.3.3 Weighing scales
- 1.3.4 Voucher collection containers and preservative
- 1.3.5 Sorting buckets and trays

1.4 Water Chemistry Measurements

- 1.4.1 In-Situ AquaTROLL 500, YSI® Professional Plus, Pro ODO
- 1.4.2 Secchi disk
- 1.4.3 Laser rangefinder

2.0 Electrofishing Survey Procedures

2.1 Training

- 2.1.1 Each ORSANCO staff member involved in electrofishing activities will be trained in electrofishing procedures by a permitted ORSANCO field crew leader.
- 2.1.2 *Field crew leader qualification* requires a biologist with supervised boat electrofishing experience, demonstrated competency with modern electrofishing equipment and techniques, demonstrated competency with fish identification and marked proficiency with SOP execution. Field crew leaders are appointed based

on eligibility, experience and professional ability as determined by and at the sole discretion of the Technical Programs Manager.

- 2.1.3 Staff shall perform at least two training sessions to the satisfaction of the Technical Programs Manager before performing any program sampling.
- 2.1.4 Each staff member involved in electrofishing activities will be certified in cardiopulmonary resuscitation (CPR) and basic first aid procedures.

2.2 Field Methods

2.2.1 Site Selection

Probabilistic sites are selected and sample frames are generated using RF3 river double lines for the Ohio River and river mile coverage provided by ORSANCO. A generalized random tessellation stratified survey design for a linear network with reverse hierarchical randomization is used to select all sampling locations. This survey design provides coordinates for 15 primary sampling sites and up to 25 overdraw sampling sites in each of the selected pools.

2.2.2 **Zone Measurement**

Standard electrofishing zones are 0.5-km length. Distances are measured with handheld GPS systems in conjunction with boat mounted chart plotters and laser rangefinders.

2.2.3 Zone Delineation

The boundaries of each electrofishing zone are clearly marked on a stationary object (e.g. trees, rocks, etc.) with biodegradable orange or red surveyor's flagging. This enables accurate location of the site on subsequent sampling dates. Care must be taken not to mark objects on private property.

2.2.4 Site Indexing

Each sampling zone location is indexed to the nearest tenth of a river mile using Geographic Information Systems (GIS) software. GPS is used to ground-truth and obtain coordinates (decimal degrees) on site at the upstream boundary of each zone.

2.3 Water Chemistry Parameters

2.3.1 Dissolved oxygen, conductivity, temperature, pH, and Secchi depth are recorded at the upstream end of each electrofishing zone prior to sampling. General weather and ambient condition observations are recorded at this time. This information is recorded at the appropriate locations on the data sheet (Attachment A).

2.4 Fish Sampling Procedures

2.4.1 Electrofishing Boat Design

A description of the electrofishing boat is given in Section 1.1

2.4.2 MLES® Infinity System Settings

The Ohio River's relative conductivity values normally range from 300 to 500 mmhos/cm. This generally results in a voltage selection of 180-225 volts DC at 60-120 pulses/ second with a duty cycle approaching 35%. These settings will generally produce the target power approaching 6000-6500W. The operator may adjust settings to produce the target power as necessary to ensure desired effect on fish. The operator may use higher voltage settings at lower conductivity readings and lower voltage settings at higher conductivity readings to obtain the desired power output.

2.4.3 Pulsed DC Electrofishing

Pulsed DC electricity is transmitted through the water by the electrode array. Safety features include a positive pressure cut-off switch located on the bow of the boat controlled by a netter and an emergency shut-down switch operated by the driver.

- 2.4.4 Surveys will be conducted at night beginning just after dusk. Night electrofishing is conducted to take advantage of increased foraging activity and diurnal movements of fishes that occur along the shoreline in the evening hours.
- 2.4.5 Individual sampling zones are electrofished from upstream to downstream by slowly and steadily maneuvering the boat close to the shore and instream structure in a "zigzag" pattern.
- 2.4.6 Time electrofished (seconds) is recorded from the MLES® control box immediately after electrofishing. A minimum of 1800 seconds is required to provide a sufficient sample. More time may be necessary for zones that incorporate more complex habitats such as those with extensive woody cover such as logs or stumps. Less time may be acceptable for zones that exhibit fast flow and/or minimal structure or instream cover.
- 2.4.7 A sampling crew consists of two netters and a driver. All personnel are clad in rubber-soled footgear, clear protective eyewear, and an orange Type I USCGA personal floatation device with reflective material. PFDs will be worn by all personnel when the watercraft is underway. The netters may also wear protective gloves.
- 2.4.8 As the driver maneuvers the boat through the electrofishing zone, netters remove affected fish from the water. The fish are then placed into a live well to be processed immediately after electrofishing.
- 2.4.9 It is recommended that sampling take place under stable, low-flow conditions at a stage level within 1m of "normal, flat pool", and when Secchi depths are at least 0.3m (13").

3.0 Fish Processing Procedures

- 3.1 Fish may be sorted into sorting containers by family or species
- 3.2 Processing priority is as follows:
 - 1. stressed individuals
 - 2. threatened and/or endangered species
 - 3. large individuals
 - 4. general population

Total length of each fish is measured to the nearest 3cm size class using a 1-meter measuring board. Total weight (when taken) of each fish is recorded to the nearest gram using either a 1.0-kg scale or a 4.0-kg scale, depending on fish weight. Small individuals of a given species may be sorted into 3-cm size classes and a total number recorded for all individuals. Large fish (>30-cm) should be measured individually, even if in large numbers. All areas of the data sheet (Figure 1) are filled out completely and legibly for each individual or size class.

4.0 Fish Disposal Procedures

- 4.1 All living specimens, except voucher and questionable specimens are returned to the water. Voucher specimens are preserved in 10% formalin solution. When handling formalin, eye protection will be worn at all times. All handling of formalin or formaldehyde will take place in a well-ventilated area. Waste formalin will be recycled for later use.
- 4.2 Fish not surviving will be buried on shore or returned to deep water for nutrient recycling. If many fish are not surviving, the project leader must investigate probable causes and implement immediate corrective action. Probable causes to be examined (but not limited to) are:
 - 1. lack of sufficient aeration
 - 2. slow / inefficient fish processing; improper fish handling
 - 3. electrofishing settings incorrect (Section 2.4.2).

5.0 Data Handling and Analysis

- 5.1 Field data sheets are physically checked, digitally photographed after each site and collected at the conclusion of each study by the Principal Investigator. All data is entered electronically as soon as possible after any field operation. Upon return, all electronic data and photo backups are moved to ORSANCO servers for QAQC and storage. Details can be found in the Data Entry and Database Usage SOP.
- 5.2 ORSANCO staff compiles and reviews all data prior to entry into ORSANCO databases. Data reduction procedures are documented in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 9.
- 5.3 For specific routine data assessment procedures, see Section 12 of the Biological Monitoring and Assessment Quality Assurance Program Plan.

6.0 Reference and Voucher Collections

Any species contained in the voucher collection but not in the reference collection will be properly labeled and added to the reference collection. Reference collections will be stored at ORSANCO.

7.0 Corrective Action

Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

8.0 References

Kolz, A.L., J.B. Reynolds, and J. Boardman. 1991. Principles and Techniques of Electrofishing. U. S. Fish & Wildlife Service Instructional Course Packet #2101

Ohio Environmental Protection Agency. 1989. Biological Criteria for the protection of aquatic life: Volume

III. Standardized Biological Field Sampling and Laboratory Methods for assessing Fish and Macroinvertebrate Communities. Division of Water Quality Monitoring and Assessment. Columbus, Ohio.

Pflieger, W.L. 1975. The Fishes of Missouri. Western Publishing Co. 343 pp.

Tennessee Valley Authority. 1987. Field Operations Biological Resources Procedures Manual. Tennessee Valley Authority, Division of Natural Resource Operations. Knoxville, Tennessee.

Trautman, M.B. 1981. The Fishes of Ohio. Revised Edition. Ohio State University Press. Columbus, Ohio. 782 pp.

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Figure 1. Electrofishing Datasheet

APPENDIX B

STANDARD OPERATING PROCEDURES

FOR

FISH TISSUE SAMPLE COLLECTION



This document describes the procedures for ORSANCO's Fish Tissue Sample Collection and provides guidelines for the proper collection, holding, sample handling and processing of Ohio River Basin Fish harvested during electrofishing activities for tissue sampling purposes. This SOP has been developed to maintain continuity and ensure data quality.

1.0 Fish Collection & Holding

Standardized electrofishing techniques are employed to collect fish for fish tissue analysis. Captured fish are held in a 50 - 75 gallon aerated live well until euthanized with a swift cervical/cranial blow. Individual fish information is collected (species, length, weight, number of individuals), and the smallest fish in each composite sample must be at least 75% of the length of the largest.

Euthanized fish will be composited whole, labeled, and each multi-fish composite doublepackaged in plastic or polyethylene bags (to minimize contamination potential between samples). A data label detailing pool, date, species, river mile, and number of fish in composite (Figure 1) will be placed between the two plastic or polyethylene bags to ensure legibility and label integrity during storage on ice, freezing and handling.

ORSANCO FISH TISSUE SAMPLE
Pool/River:
Date:
Species:
Rmi: OBP
Fish: of

Figure 1. Example fish tissue sample data label.

While stored on wet ice, make certain sufficient ice comes in contact with the composite sample to maintain ~4 C temperature. If stored with dry ice, make certain the sample comes in contact with the dry ice to ensure freezing. Transport all samples to the ORSANCO offices as soon as possible for storage in designated freezers until they are shipped frozen (whole) to the contract analytical laboratory.

2.0 Shipping

Whole-fish composite samples may be shipped to the contract laboratory throughout the field season whenever feasible and no later than 11/15 to ensure timely analysis data receipt. Frozen composites are placed in a large plastic bag, sealed (zip tie), and placed in coolers with accompanying chain of custody (COC) documentation. The COC will also be emailed to the contract laboratory prior to shipping. Sample coolers will be shipped overnight to the contract laboratory no later than Thursday to ensure they are received prior to a weekend and remain frozen until processed.

3.0 Sample Processing

Fish will be thawed, weighed, measured, and filleted / mechanically separated at the contract laboratory prior to grinding and analysis. All sample metadata will be catalogued and inventoried in spreadsheets by ORSANCO staff prior to receiving length, weight and analysis data from the contract laboratory.

4.0 Data Entry and Handling

The contract laboratory will deliver all analysis and supporting documentation to ORSANCO in electronic format unless otherwise requested upon completion. Data entry and database usage procedures are outlined in ORSANCO's Quality Assurance Program Plan for the Biological Monitoring and Assessment Program Appendix I: Standard Operating Procedures for Data Entry and Database Usage.

5.0 Corrective Action:

Issues or potential improvements pertaining to the collection of fish tissue may arise during the execution of this protocol. Corrective actions may therefore be implemented at the discretion of the crew leader. Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

APPENDIX C

STANDARD OPERATING PROCEDURES

FOR

HABITAT DATA COLLECTION FOR

FISH POPULATION SURVEYS



This document describes the procedures for ORSANCO's Habitat Data Collection. This SOP has been developed to maintain continuity and ensure data quality.

1.0 Field Equipment

The following equipment is used to conduct the habitat surveys:

1.1 Watercraft

- 1.1.1 19-ft. aluminum johnboat
- 1.1.2 Two 10-ft ³/₄ in. copper poles capped at one and wrapped with marking tape at each one foot interval along the length of the pole. Poles are fitted with male and female adapters respectively.
- 1.1.3 Laser rangefinder
- 1.1.4 Marine radio

2.0 Habitat Data Collection Procedures

2.1 Training

- 2.1.1 Each ORSANCO staff member involved in the habitat data collection program will be trained in collection procedures by a staff member having at least one year of habitat data collection experience on the Ohio River.
- 2.1.2 *Field crew leader qualification* requires a biologist with supervised boat experience, demonstrated competency with modern equipment and techniques, and marked proficiency with SOP protocol execution. Field crew leaders are appointed based on eligibility, experience and professional ability as determined by and at the sole discretion of the Technical Programs Manager.
- 2.1.3 Staff shall perform at least two training sessions to the satisfaction of the Crew Leader and Technical Programs Manager before performing any program sampling.

2.2 Field Methods

2.2.1 Site Selection

Sites are selected and sample frames are generated using RF3 river double lines for the Ohio River and river mile coverage provided by ORSANCO. A generalized random tessellation stratified survey design for a linear network with reverse hierarchical randomization was used to select all sampling locations. This survey design provided coordinates for 15 primary sampling sites and up to 25 overdraw sampling sites in each of the selected pools.

2.2.2 Zone Measurement

Electrofishing zones are 0.5-km length. Distance is measured with a handheld GPS unit and a laser rangefinder. Sampling Zones are measured by navigating to the top or upstream end of the electrofishing zone, marking the start point in the GPS unit while simultaneously measuring distance from shore with the laser rangefinder. These two pieces of equipment are used to measure the length of the zone and distance from shore while slowly maneuvering the boat to the end of the zone.

2.2.3 Zone Delineation

The boundaries of each electrofishing zone are clearly marked on stationary object (e.g. trees, rocks, etc.) with florescent orange paint and/or orange surveyor's flagging. This enables accurate location of the site on subsequent sampling dates. Care must be taken not to mark objects on private property.

2.2.4 Site Indexing

Each sampling zone location is indexed to the nearest tenth of a river mile using Geographic Information Systems (GIS) software. GPS is used to ground-truth and obtain coordinates (decimal degrees) on site at the upstream boundary of each zone.

2.3 Habitat Data Collection Procedure

2.3.1 Beginning at shoreline at the upstream end of the zone, one crew member takes/ records a GPS mark at the water's edge. Sediment at the shoreline is recorded. The driver then slowly backs the boat away from shore in a straight line perpendicular to the shoreline, as a crewmember maintains a fix on the target point with the rangefinder and calls off distance to shore. At each 10' interval sediment and depth are recorded by lowering one end of the copper pole to the substrate. Sediment and depth are recorded every 10' to 100' out from shore. This procedure is repeated at each of the six 100 m marks of the zone, starting at the upper end and finishing at the lower.

3.0 Data Handling and Analysis

Field data sheets are checked and initialed by at least two crewmembers, digitally photographed and are collected at the conclusion of each study by the Principal Investigator. Details can be found in the Date Entry and Database Usage SOP. ORSANCO staff compiles and reviews all data prior to entry into the ORSANCO data base.

4.0 Corrective Action

4.1 Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

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APPENDIX D

STANDARD OPERATING PROCEDURES

FOR

AQUATIC VEGETATION COLLECTION FOR

FISH POPULATION SURVEYS



This document describes the procedures for ORSANCO's Aquatic Vegetation Collection. This SOP has been developed to maintain continuity and ensure data quality.

1.0 Field Equipment

The following equipment is used to conduct the aquatic vegetation surveys:

1.1 Watercraft

- 1.1.1 19-ft. aluminum boat
- 1.1.2 Doubled-headed vegetation rake
- 1.1.3 Laser Rangefinder
- 1.1.4 Marine radio

2.0 Aquatic Vegetation Collection Procedures

2.1 Training

- 2.1.1 Each ORSANCO staff member involved in the aquatic vegetation data collection program will be trained in collection procedures by a staff member having at least one year of habitat data collection experience on the Ohio River.
- 2.1.2 *Field crew leader qualification* requires a biologist with supervised boat experience, demonstrated competency with modern equipment and techniques, and marked proficiency with SOP protocol execution. Field crew leaders are appointed based on eligibility, experience and professional ability as determined by and at the sole discretion of the Technical Programs Manager.
- 2.1.3 Staff shall perform at least two training sessions to the satisfaction of the Crew Leader and Technical Programs Manager before performing any program sampling.

2.2 Field Methods

2.2.1 Site Selection

Sites are selected and sample frames are generated using RF3 river double lines for the Ohio River and river mile coverage provided by ORSANCO. A generalized random tessellation stratified survey design for a linear network with reverse hierarchical randomization is used to select all sampling locations. This survey design provides coordinates for 15 primary sampling sites and up to 25 overdraw sampling sites in each of the selected pools.

2.2.2 Zone Measurement

Electrofishing zones are 0.5-km length. Distance is measured with a handheld GPS unit and a laser rangefinder. Sampling Zones are measured by navigating to the top or upstream end of the electrofishing zone, marking the start point in the GPS unit while simultaneously measuring distance from shore with the laser rangefinder. These two pieces of equipment are used to measure the length of the zone and distance from shore while slowly maneuvering the boat to the end of the zone.

2.2.3 Zone Delineation

The boundaries of each sampling zone are clearly marked on stationary object (e.g. trees, rocks, etc.) with florescent orange/ red surveyor's flagging. This enables accurate location of the site on subsequent sampling dates. Care must be taken not to mark objects on private property.

2.2.4 Site Indexing

Each sampling zone location is indexed to the nearest tenth of a river mile using Geographic Information Systems (GIS) software. GPS is used to ground-truth and obtain coordinates (decimal degrees) on site at the upstream boundary of each zone.

2.3 Aquatic Vegetation Data Collection Procedure

- 2.3.1 Beginning at the shoreline at the upstream end of the zone, one crew member takes/ records a GPS mark at the water's edge, marking the top of the zone (0m). Five transects are marked in 100m intervals downstream of 0m. Between each of these six transects visual methods are used to provide a qualitative estimate of woody cover, submerged, and emergent vegetation occurrence between each transect. The driver then slowly backs the boat away from shore in a straight line perpendicular to the shoreline, as a crewmember maintains a fix on the target point with the rangefinder and calls off distance to shore. Aquatic vegetation is gathered at each 10' interval using a double sided rake. In water shallower than 15 feet deep, a rake attached to a pole is lowered to the substrate. The rake is then twisted around twice and pulled straight out of the water. This procedure is repeated at each of the six transects throughout the zone (0m, 100m, 200m, 300m, 400m, 500m). The plants collected at each transect will be composited at completion of the zone. Once separated, the total biomass will be measured for each species.
- 2.3.2 Data recorded at each transect point include:
 - 2.3.2.1 Sampling device 'P' pole, 'R' rope
 - 2.3.2.2 Rake fullness '0' No plants present, '1' Only a few plants present, not enough to cover entire length of rake, tines still visible, '2'– There are enough plants to cover entire length of rake in a single layer, tines not fully covered, '3' the rake is completely covered in plants, tines not visible. These measures are obtained for each species observed on the rake. Plants that are dislodged via the rake but either fall off or float to surface are included in fullness measures. When a species is observed in the vicinity of a sample point, within 5 feet, but not sampled by the rake, the species is recorded as visually observed and included in total number of species observed.
 - 2.3.2.3 Species name Taxa codes for each species will be recorded.
 - 2.3.2.4 **Voucher type** 'P' photo, 'S' specimen
 - 2.3.2.5 Species Biomass Biomass (grams) will be recorded for each species

3.0 Data Handling and Analysis

- 3.1 Field data sheets are checked and initialed by at least two crewmembers, digitally photographed and are collected at the conclusion of each study by the Principal Investigator. Details can be found in the Date Entry and Database Usage SOP.
- 3.2 ORSANCO staff compiles and reviews all data prior to entry into the ORSANCO data base.

4.0 Corrective Action

4.1 Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

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100	30	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
100	40	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
100	50	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
100	60	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
100	70	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
100	80	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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200	20	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
200	20		0 1 2 2	0 1 2 2	0 1 2 3	0 1 2 2	0 1 2 2	0 1 2 3	0 1 2 3
200	40		0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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200	80	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
200	90	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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Figure 1. ORSANCO Aquatic Vegetation Field Sheet – Front.

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300	20	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	30	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	40	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	50	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	60	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	70	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
300	80	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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400	10	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
400	20	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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400	50	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
400	60	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
400	70	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
400	80	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
400	90	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
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500	80	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
500	90	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
500	100	PR	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3	0 1 2 3
	Vou	cher Type	Photo Sample	Photo Sample	Photo Sample	Photo Sample	Photo Sample	Photo Sample	Photo Sample
			oumplo	ounplo	Gample		Gampio	canpio	ounplo
Visua	Veg. Ob	servation:			-	-	-		
				Sp	ecies Name (a	bbreviate & re	cord total biom	ass)	
	Specie	Diamage	-	_	-	-	_	_	_
	Species	s biomass:	g	g	g	g	g	g	g

Figure 2. ORSANCO Aquatic Vegetation Field Sheet – Back.

Fullness Rating	Coverage	Description
0		No plants present.
1	King and a start fring	Only few plants. There are not enough plants to entirely cover the length of the rake head in a single layer.
2	A Property and	There are enough plants to cover the length of the rake head in a single layer, but not enough to fully cover the tines.
3		The rake is completely covered and tines are not visible.

Figure 3. Illustration of rake fullness ratings modified from Hauxwell et al. 2010.

APPENDIX E

STANDARD OPERATING PROCEDURES

FOR

MACROINVERTEBRATE SAMPLING

USING MODIFIED HESTER-DENDY SAMPLERS (Deep Water Version)



This document describes the procedures for ORSANCO's aquatic macroinvertebrate population surveys using the modified Hester-Dendy (H-D) multi-plate sampling method.

1.0 Sampling Schedule

Sampling schedules should be established which take advantages of low flow conditions in summer and early fall. The samplers are set out in late August to mid-September, and are collected six weeks after placement. (Attachment 1, 2017 Intensive Survey Sampling Workplan)

1.1 **Personnel Qualifications**

Field crew leader qualification requires a biologist with supervised boat experience, demonstrated competency with modern equipment and techniques, and marked proficiency with SOP protocol execution. Field crew leaders are appointed based on eligibility, experience and professional ability as determined by and at the sole discretion of the Technical Programs Manager.

2.0 Sampling Procedures

2.1 Hester-Dendy (H-D) Specifications

Samplers are constructed of 1/8 inch tempered masonite cardboard cut into three inch square plates and one inch square spacers. A 3/8 inch hole is drilled in the center of each plate and spacer. Eight plates and twelve spacers are placed on a 1/4 inch X 4 inch eye bolt such that there are three single spaces (1/8"), three double spaces (1/4"), and one triple space (3/8") between the plates. Plates and spacers are secured to the eye bolt with two 1/4 inch washers and one standard 1/4 inch nut. See Hester and Dendy (1962).

2.2 Sampling Unit Assembly

A sampling unit is a series of five H-D samplers bound together with twine or cords and secured to a cement block. The five samplers are tied together, eyebolt to eyebolt in a circular pattern. The group is then lashed securely to the top of the block with cord of at least 1/8" diameter. A two-foot piece of reinforcing rod is secured vertically to the block to be partially driven into the substrate for additional stability.

2.3 Placement

Each sampling unit should be placed in an area safe from disturbance in substrate representative of the 500m zone. In the event that the zone is not suitable for deployment, the field crew leader may then choose to set in a nearby area that would best represent the zone, ideally selecting a location within the site. Once a location is chosen, a boat driver backs out slowly from shore, until 10' depth is achieved. The sampling unit is lowered into the water by the line and allowed to settle on the bottom. The boat then returns to shore, letting line out so that it may be tied off securely. The line may be tied to anything deemed secure by the collector. Efforts will made to disguise the line to prevent vandalism.

2.4 Colonization period

The sampling unit must remain undisturbed for a period of at least six weeks, but should not exceed eight weeks.

2.5 Sampling Unit Retrieval

During retrieval, the sampling unit is approached from downstream to ensure minimal disturbance. Upon location of the retrieval line, the collector will cut the line, being sure to keep the line taught to minimize disturbance. The collector then backs out slowly until the boat is directly over the sampling unit, at which time the unit is slowly pulled to the surface. A five-gallon bucket is submerged and positioned next to the unit. The five H-D samplers are then carefully cut from the block and slid into the bucket. The bucket is then taken to the boat and the plates disassembled.

2.6 Plate Disassembly and Sample Preservation

The five H-D samplers are disassembled in the bucket with special care taken not to spill or lose any of the sample material. The plates are brushed or scraped using another plate while submerged and all sampler parts rinsed with distilled water and discarded. The bolts may be kept for reuse. After all parts have been rinsed and removed from the bucket, the water is then poured through a standard #30 sieve, the bucket is rinsed through the sieve until clean and all residue placed in a sample container. The sieve is rinsed repeatedly into a white sorting pan or bucket to ensure that all organisms have been removed from the sieve. Once all organisms and residue are in the sample container, 10% formalin is added to cover the sample with at least one inch of preservative.

2.7 Sample Packaging and Labeling

Each sample is properly preserved in a plastic sample container. The lid is then sealed shut with electrical tape and labeled. Each container is labeled with collection site, date of collection, sample number and GPS coordinates. An additional tag made of waterproof paper and permanent ink is placed in the jar. All samples are recorded on a standard chain of custody form (Figure 1).

2.8 Sample Storage

Preserved samples are held at ORSANCO at room temperature until they can be shipped to the lab.

2.9 **Documentation**

Habitat and environmental conditions, such as water quality parameters at each sampling location are noted and recorded. A standard macroinvertebrate-sampling sheet is used to record the locations of sampler placement and retrieval.

3.0 Corrective Action

Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

4.0 Materials List

PLACEMENT:

- 1. Waders
- 2. Assembled H-D samplers
- 3. Paver stone

- 4. 2 ft. piece of rebar
- 5. Rope
- 6. Hammer

RETRIEVAL:

- 1. Waders
- 2. Knife
- 3. 5-gallon bucket
- 4. Crescent wrench
- 5. Common screwdriver
- 6. Squirt bottle
- 7. Distilled water
- 8. Sorting pans or buckets
- 9. #30 sieves
- 10. Plastic sample jars and lids
- 11. Electrical tape
- 12. Permanent markers
- 13. Waterproof paper and ink for labels in sample jars
- 14. Any instruments needed for measuring WQ parameters

5.0 Literature Cited

Hester, F. E., and J. S. Dendy. 1962. A multiple-plate sampler for aquatic macroinvertebrates. *Transactions of the American Fisheries Society* 91:420–421.

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Figure 1. Example chain of custody form

APPENDIX F

STANDARD OPERATING PROCEDURES

FOR

MACROINVERTEBRATE SAMPLING USING A QUALITATIVE MULTIPLE HABITAT APPROACH



This document describes the procedures for ORSANCO's aquatic macroinvertebrate population surveys using the qualitative multiple habitat sampling method.

1.0 Sampling Schedule

The qualitative sampling methods are to be performed upon retrieval of the Hester-Dendy sampling units.

2.0 Sampling Procedures

2.1 Net Specifications

Samples are collected with standard D-frame 500µm mesh dip nets.

2.2 Collecting Technique

Multihabitat samples are collected at 6 transects, every 100m, throughout each 500m zone. At each transect, 10 of any combination of jabs, sweeps, kicks, etc. are taken within 10m of the transect point. Efforts should be made to sample all available habitats within the 10m radius. The net is rinsed of debris and organisms into a bucket at each transect, with all transects being combined to make one composite sample.

2.3 Plate Disassembly and Sample Preservation

After all 6 transects have been collected, the remaining slurry is then poured through a standard #30 sieve, the bucket is rinsed through the sieve until clean and all residue placed in a sample container. The sieve is rinsed repeatedly into a white sorting pan or bucket to ensure that all organisms have been removed from the sieve. Once all organisms and residue are in the sample container, 70% ethanol or 10% formalin (as required by contractor) is added to cover the sample with at least one inch of preservative.

2.4 Sample Packaging and Labeling

Each sample is preserved in a plastic sample container. The lid is sealed shut with electrical tape and labeled. Each container is labeled with collection site, date of collection, and sample number. An additional waterproof paper tag is filled out (no. 2 pencil) and placed in the jar. All samples are recorded on a chain of custody form (Figure 1).

2.5 Sample Storage

Preserved samples are held at ORSANCO at room temperature until they can be shipped to the lab.

2.6 **Documentation**

Habitat and environmental conditions, such as water quality parameters at each sampling location are noted and recorded in a log. A standard macroinvertebrate-sampling sheet is used to record the locations of sampler placement and retrieval.

3.0 Corrective Action

Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by crew leaders and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

4.0 Materials List

- 1. Waders
- 2. D-frame net
- 3. Buckets
- 4 Squirt bottles
- 5. Distilled water
- 6. Sorting pans or buckets
- 7. #30 sieves
- 8. Plastic sample jars and lids
- 9. Electrical tape
- 10. Permanent markers
- 11. Waterproof paper and ink for labels in sample jars
- 12. Any instruments needed for measuring WQ parameters

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Figure 1. Example chain of custody form

APPENDIX G

STANDARD LABORATORY PROCEDURES

FOR THE

MACROINVERTEBRATE SAMPLE ANALYSIS



ORSANCO has adopted a version of Ohio EPA's laboratory methods for the analysis of its macroinvertebrate samples. Our methods are based Ohio EPA document: Biological Criteria for the Protections of Aquatic Life: Volume III. "Standardized biological field sampling and laboratory methods for assessing fish and macroinvertebrate communities". Ohio Environmental Protection Agency. 1989. Division of Water Quality Monitoring an Assessment, Columbus, Ohio.

1.0 Laboratory Procedures

Macroinvertebrate samples are immediately given an acceptable identification code and recorded into a log book upon arrival at the laboratory. Other information in the logbook includes the sample location, retrieval date, the person who conducts the analysis, and any other comments considered pertinent to the collections and sample analysis.

Composite samples are passed through U.S. Standard Testing Sieves #30 (0.589mm openings) with all remaining material preserved and properly labeled and coded in containers of 70% ethanol.

The following procedures are used during sample analysis:

- A) Sorting of the sample is done in a white enamel pan followed by scanning under the dissecting microscope (10x magnification). Subsamples are produced using the following guidelines:
 - 1. The Folsom sample splitter is used for all subsampling.
 - 2. After an entire sample has been sorted, subsampling within families containing unmanageable numbers is acceptable.
 - 3. Very large samples may be subsampled prior to sorting but only after examination in a white enamel pan to remove obvious rare taxa, e.g. crayfish, hellgrammites, non-hydropsychid caddisflies.
 - 4. A minimum of 250 organisms is identified, with at least 50-100 midges, 70 caddisflies, 70 mayflies.
- B) Dipterans of the family Chironomidae are prepared for identification by clearing the larvae in hot 10% KOH for 30 minutes and then mounting in water on a microscope slides. Permanent slides for the voucher collection are mounted in Euparol mounting medium.
- C) Organisms determined to be dead at the time of collection are discarded.
- D) When only one sex of life stage can be identified it is assumed that the other sex or stage is the same species.
- E) Sections of bryozoan colonies are removed from the plates and saved for identification. Only colonies, not individuals, are counted.
- F) Early instars that cannot be identified are extrapolated where possible.
- G) Species level identifications are made wherever possible and practical. Generic or higher level classifications are made if specimens are damaged beyond identification, in those cases where taxonomy is incomplete or laborious and time-consuming, or where the specimen is an unidentifiable early instar.
- H) Organisms are listed in a standard laboratory table format.
- I) Two end fragments of an oligochaete are counted as an individual. Fragments without ends are not counted.

J) Any taxonomic key in the laboratory can be used as an aid in the identification of an organism. However, the final identification and name used are taken from the asterisked references in the attached tables. Also indicated is the level of taxonomy attainable with the keys listed.

2.0 Corrective Action

Issues or potential improvements pertaining to the analysis of macroinvertebrate samples may arise during the execution of this protocol. Corrective actions may therefore be implemented at the discretion of ORSANCO Biological Staff. Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by the Project Leader and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

APPENDIX H

STANDARD OPERATING PROCEDURES

FOR

DATA ENTRY AND DATABASE USAGE



This document describes the procedures for ORSANCO's biological data entry and database use. This document provides guidelines for the proper entry, archiving and processing of Ohio River Basin location, fish, macroinvertebrate, habitat, water quality, and background data collected during monitoring and assessment activities. This SOP has been developed to maintain continuity and ensure the quality of the data collected and subsequent products derived from these data.

1.0 Data Entry Schedule

Data entry occurs throughout the field season once data have been collected and all data sheets are returned to ORSANCO. Database architecture, appending and verification occur at the cessation of field activities.

2.0 Data Entry Procedures

Raw data sheets are assembled in a file along with site description sheets, maps of the sampling sites, and the final study plan. Raw data are entered by biological staff under the supervision of the Project Leader into Microsoft Excel© spreadsheets and imported into a Microsoft Access© relational database under the supervision of the Project Leader. Data are to be manually entered sequentially row by row from the raw datasheet specific to each location. Data entry will continue for all data types until each datasheet is initialed as complete. Any changes made to data sheets are initialed, dated and verified by the Project Leader. Digital photograph backups of all datasheets will be compiled, organized by date and location and stored electronically on ORSANCO servers.

After all data for a survey have been entered into the database, the entries are proofread by the Field Sampling Leader for accuracy; a final call on whether the data is acceptable will be made by the Project Leader. All corrections or updates are then applied to the database.

3.0 Database Usage

The biological Microsoft Access[©] database(s) will reside and be maintained on ORSANCO servers and their security and maintenance overseen by the ORSANCO IT staff.

Access to these data servers is possible only by ORSANCO staff using a valid login and password. All data on all servers are backed up incrementally on a daily basis as changes / modifications occur. Additional full-system backups occur weekly and are overseen by ORSANCO IT staff. Additionally an emergency backup of all biological database information is maintained on an external hard drive. This emergency backup is updated incrementally as changes are made to the database. The emergency backup is overseen and maintained by ORSANCO biological staff under the supervision of the Technical Programs Manager.

All databases are comprised of tables containing both text and numeric information. Queries can be performed by any member of ORSANCO biological staff with a valid login and password or any staff member authorized by and under supervision of ORSANCO staff.

Data queried from databases will be used in numerous statistical analyses tailored to provide insight as they relate to specific projects at the discretion of ORSANCO biological staff.

4.0 Database Verification

Once the database(s) is (are) populated / appended it is verified for accuracy by the Project Leader. Data tables are sorted to identify and correct clerical errors in taxonomy, location information, location naming convention, river mile, and collection information. A minimum of 10% of all data in each database is hand verified by comparison to raw datasheets. If more than 10% of the verifications are incorrect, 100% of digitally entered data are verified against original raw datasheets to ensure correct entry.

5.0 Documentation

Any and all data, documents / products, or reports derived from analyses of data within ORSANCO databases are available to the public via written request only after QAQC and review. The use of Microsoft Office© tools (Access, Excel etc.) will be documented upon request. QAQC responsibility for data entry and database usage/ verification as related to the Biological Monitoring and Assessment Program effectively terminates at the annual compilation of a final, verifiably accurate and up to date database for use by ORSANCO biological staff.

6.0 STORET Data Submission

As per Clean Water Act Section 106 funding requirements, ORSANCO will be uploading all applicable monitoring data to STORET. STORET refers overall to "STORage and RETrieval", an electronic data system for water quality monitoring data developed by EPA. Since about 2000, STORET has referred to a local data management system ("Modernized STORET") as well as data repository ("STORET Data Warehouse") developed for purposes of assisting data owners manage data locally and share data nationally. As of September 2009, the Water Quality Exchange, or WQX framework, provides the main mechanism for submitting data to the STORET Data Warehouse. ORSANCO's Biological Monitoring and Assessment Program is exploring options for data submission to STORET.

7.0 Corrective Action

Issues or potential improvements pertaining to data entry and database usage may arise during the execution of this protocol. Corrective actions may therefore be implemented at the discretion of the Project Leader. Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by staff and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

APPENDIX I

STANDARD OPERATING PROCEDURES

FOR

FIELD SAMPLING SAFETY

ELECTROFISHING CONSIDERATIONS

EMERGENCY GUIDELINES AND REPORTING



This document describes ORSANCO's Field Sampling Safety procedures, special safety considerations for electrofishing activities, injury and emergency guidelines and reporting protocol.

1.0 Training

All crew members will receive training by an ORSANCO staff member with at least three years of large river electrofishing and field sampling experience as a crew leader. All crew members will be responsible for familiarizing themselves with all applicable ORSANCO SOPs related to their workload. Crew training will take place in accordance with procedures outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 4.

2.0 General Guidelines

- 1. All crew members will wear orange USCG-approved PFDs with reflective material when on any ORSANCO watercraft that is underway.
- The locations of all on-board safety systems will be reviewed with all crew members prior to all field operations. All crew members will be advised of the on-board location and operation of cell phones, VHF radios, first aid kits, allergy pen kits, AEDs, fire extinguishers, flares, illumination devices, GPS navigation systems and vehicle keys.
- 3. Crew members will receive CPR, First-Aid, AED and safe boating training. Crew members will be trained on watercraft operation, navigation and troubleshooting to ensure that all crew members can safely and competently operate ORSANCO watercraft in the event of emergency.
- 4. All crew members will be advised of an emergency contact procedure prior to any field operations. In the event of a medical emergency ORSANCO management (HR, Program and Safety) must be notified as soon as possible as must any contracting organization that employs interns and other crew members (Section 6.0).
- 5. The Ohio River and many of its larger tributaries are subject to heavy barge and recreational boating traffic. Extreme caution must be used when navigating amongst other watercraft and floating debris. When navigating after dark, navigational running lights and a high-power spotlight are required so that other vessels are aware of the watercraft and the driver can more easily detect obstacles in the water.
- 6. Appropriate eye protection that meets or exceeds ANSI Z87.1-2015 High Velocity Impact Standards must be worn at all times when the watercraft is underway.
- 7. Large (>10 kg) Silver Carp (*Hypophthalmichthys moltrix*) can propel themselves of the water to heights exceeding 3 meters. Serious injuries have occurred due to Silver Carp collisions. Silver Carp are present throughout most of the Ohio River below McAlpine Locks and Dam. All crew members must monitor for and be aware of Silver Carp while watercraft are underway. As such, crew members will be provided protective mouth guards.
- Prior to any watercraft field operation, crew leaders will conduct a review of river conditions and stage reports as well as weather patterns and local radar. Likewise, operators will have access to up to date listings of ramps, towns, facilities and USACOE Corps Charts.
- 9. Prior to all field activities, all appropriate state entities will be notified and advised of the general sampling plan for a given week including general location, activity type and contact phone #. Additionally a plan of communication between crews and office personnel will be established.

3.0 Electrofishing Guidelines

1. Prior to each sampling event, all electrical safety switches will be checked to ensure they are working properly.

- 2. All electrical connections will be checked prior to use to ensure that they are free of corrosion and proper, tight connections are maintained.
- 3. Anodes and cathodes (including cathodes other than the hull) are not to be touched while an electrofishing generator is running. Do not reach for or touch objects outside the watercraft. Do not reach into the water outside the watercraft unless all electricity to the water has been turned off by ensuring that all switches are in the "OFF "position (generator, EF control unit, pedal).
- 4. All crew members will wear rubber soled footwear while electrofishing.
- 5. Hearing protection, eye protection, gloves and protective mouth guards will be available for all crew members.
- 6. All watercraft operations during electrofishing are to be conducted by an ORSANCO staff crew leader.
- 7. During electrofishing, the watercraft operator maintains the electrofishing unit safety switch at all times.
- 8. All crew members will be instructed on safe and proper use of long-handled electrofishing dip nets.
- 9. Do not electrofish in high waves, extreme velocity or other conditions that may cause sudden motions of the watercraft that can cause crew members to lose their balance.
- 10. Electrofishing will be conducted only when atmospheric and river conditions permit. Excessive wind, waves, rain, fog, etc. are examples of conditions that will be considered prior to initiation of an electrofishing survey. Do not electrofish when lightning is in near proximity. Survey suspension/ delay / postponement is at the sole discretion of the Crew Leader.
- 11. All engine and electrofishing fuel systems and fuel tanks will be checked and topped off, as appropriate, prior to the initiation of an electrofishing event. The on-board generator will not be refueled until such time as it has cooled sufficiently to prevent spontaneous combustion of spilled fuel. On-board fuel tanks and auxiliary fuel / engine oil cans are to be installed and stored far from heated exhaust.
- 12. All equipment, gear, supplies, personal effects, etc. will be kept clear of generator exhaust systems to prevent thermal damage or fire.
- 13. Good line of sight and verbal communication must be maintained among crew members at all times. Generators are loud and often drown out verbal communication. Hand signals should be predetermined and used to communicate direction, power on/off, and other vital information.
- 14. Driver's licenses and ID cards will be in the possession of each crew member. Scientific Collecting permits and applicable licenses will be in the possession of each crew leader at all times while conducting sampling activities.

4.0 Injury and Emergency Guidelines

- Always be vigilant of each crew member and their condition. This includes hydration, nutrition, sunscreen application, proper safe and field-appropriate attire, etc. In the event of an injury, the injured crew member must immediately notify the rest of the crew if able. If one crew member notices another is either injured or otherwise imperiled they are to immediately notify the remainder of the crew.
- As all crew members will be certified in CPR, First Aid, and AED usage, assess whether the injury can be resolved on-site using on-board kit(s), and move quickly to stabilize the injury / crew member.

- 3. In the event of serious injury or dangerous situation (weather, river conditions, driving conditions etc.) the crew must move to the closest safe location as soon as possible to expedite care for injured crew member(s) and ensure that the crew is not jeopardized.
- 4. In the event of waterborne emergencies, the crew must maneuver to shore as quickly and safely as possible while notifying local authorities of their location and emergency specifics. Follow all instructions per the 911 dispatch operator and determine a safe and feasible rendezvous point with first responders. Likewise with traffic / driving emergencies, observe all traffic regulations and ensure that the crew is met by first responders in a safe expedient manner.
- 5. As soon as possible, ORSANCO HR and the Technical Programs Manager are to be notified of any injury, traffic incident, equipment loss or damage and emergency narrative and resolution.

5.0 Injury Reporting

- 1. If the injury exceeds first aid treatment, once treated, the ORSANCO HR Director will advise the injured person and facilitate notification to the appropriate care provider that a work-related injury has occurred. Additionally, the injured person must notify the care giver that Sheakley is the managed care organization (MCO). This is key to receiving prompt care and claim processing.
- Within 24 hours of receiving treatment the injured individual must complete a BWC First Report of Injury Form (FROI-1) and submit to the ORSANCO HR Director for processing (management will provide and assist in completion of this form).
- 3. The form will be transmitted to Sheakley along with treatment documentation if available.
- 4. The ORSANCO HR Director will review and track the workers' compensation claim.
- 5. As soon as possible after the occurrence of an on-the-job injury or accident, the crew leader(s) will investigate the incident and also immediately notify the ORSANCO HR Director and the Technical Programs Manager. An investigation report will be completed and filed with the ORSANCO HR Director as appropriate.

6.0 Corrective Action

Issues or potential improvements pertaining to ORSANCO sampling / watercraft safety may arise during the execution of this protocol. Corrective actions may therefore be implemented at the discretion of the Project Leader and ORSANCO Safety Officer. Immediate Corrective Actions (ICAs) and Long Term Corrective Actions (LTCAs) will be determined by staff and documented as per the procedure outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 13.

APPENDIX J

STANDARD OPERATING PROCEDURES

FOR

MULTI-PARAMETER WATER QUALITY SENSOR

DEPLOYMENT AND APPLICATION



This document describes ORSANCO's Multi-parameter Water Quality Sensor (i.e., continuous data loggers) deployment, application and data handling procedures.

1.0 Field Equipment

The following equipment is used to deploy Multi-parameter Water Quality Sensors:

1.1 Multi-parameter Water Quality Sensor Unit Types

- 1.1.1 Onset HOBO® Dissolved Oxygen/Temperature Data Logger
- 1.1.2 YSI 6600V2
- 1.1.3 In-Situ AquaTROLL 500
- 1.1.4 YSI EXO2

1.2 Watercraft (where applicable)

- 1.2.1 Approved workboat equipped with dual engines and all appropriate safety gear
- 1.2.2 Handheld GPS
- 1.2.3 Laser rangefinder / measuring tape
- 1.2.4 Marine radio
- 1.2.5 Tool kit
- 1.2.6 Data sheets / field notebook

1.3 Sensor Mount / Sampling Platform and Security

- 1.3.1 PVC protective sheath
- 1.3.2 Parachute (550) cord (in stream) or steel cable (infrastructure-attached sites)
- 1.3.3 Concrete cinder block, heavy grade zip-tie fasteners, 2' rebar sections

2.0 Training

- 2.1 All crew members will receive training by an ORSANCO staff member with at least one year of large river field sampling experience as a Field Crew Leader. All crew members will be responsible for familiarizing themselves with all applicable ORSANCO SOPs related to their workload. Crew training will take place in accordance with procedures outlined in the Biological Monitoring and Assessment Quality Assurance Program Plan, Section 4. All field sampling will occur under the direct supervision of an ORSANCO Field Crew Leader.
- 2.2 *Field crew leader qualification* requires a biologist with supervised boat experience demonstrated competency with modern equipment and techniques, and marked proficiency with SOP protocol execution. Field crew leaders are appointed based on eligibility, experience and professional ability as determined by and at the sole discretion of the Technical Programs Manager.
- 2.3 Staff shall perform at least one training session to the satisfaction of the Field Crew Leader and Technical Programs Manager and be familiar with all ORSANCO Field Safety protocols before performing any program sampling.

3.0 Field Methods for Multi-parameter Water Quality Sensor Deployment

3.1 Sensor Initialization, Calibration and Sampling Platform Construction

Each Multi-parameter Water Quality Sensor will be initialized and indexed prior to deployment using instrument-specific software packages. Once initialized, each sensor will be prepared as per manufacturer guidelines and carefully packed for transport to the

deployment site(s). Sensors that are factory sealed require calibration by the manufacturer, and are shipped back for inspection and calibration within the manufacturer-specified timeframes. All other water quality sensors will be calibrated every two weeks. This calibration will be conducted in accordance with all manufacturer's instructions and guidelines. Regardless of type, all sensors are returned to the manufacturer for routine maintenance within the manufacturer-specified timeframes.

Upon deployment, sensors are unattended and deployment sites are of two types:

-in-situ in stream - or -in-situ infrastructure-attached.

Each in-situ in stream deployment for Onset HOBO® data loggers consists of:

- 3.1.1 1 (one) full size foundation-grade concrete cinder block and 2' rebar sections
- 3.1.2 1 (one) 2" PVC sheath and cap; each sheath drilled with a sufficient number of 0.5" holes to allow adequate water flow around the sensor
- 3.1.3 1 (one) water quality sensor
- 3.1.4 adequate parachute cord to secure sensor to sheath cap (cap is attached to PVC sheath via friction) and to onshore tie-off location (e.g., tree or other fixed object)

In-situ in-stream deployment illustration



Each in-situ infrastructure-attached deployment for Onset HOBO® data loggers consists of:

- 3.1.5 1 (one) 2" PVC sheath drilled with a sufficient number of 0.5" holes to allow adequate water flow around the sensor
- 3.1.6 1 (one) water quality sensor
- 3.1.7 heavy duty cable tie (for conductivity sensors) or bolt with wing nut to secure sensor to PVC sheath
- 3.1.8 adequate steel cable to secure sheath to attached infrastructure for security and retrieval
- 3.1.9 lead or concrete weight attached to bottom of sheath via steel cable

Each PVC protective sheath must be sufficiently robust to handle the impact of large, fast-moving debris flowing with the water.



In-situ infrastructure deployment illustration

3.2 Site Selection and Equipment Location Considerations

Exact sampling sites will change periodically and will be documented in annual work plans.

3.2.1 In-situ in stream water quality sensor deployment site locations that are coupled with existing monitoring / assessment site locations should be placed within the sampling reach, preferably near the downstream end. Care should be taken in choosing exact deployment sites. Areas of potentially high human activity (fishing areas, boat docks, etc) should be avoided to minimize tampering with the sensors. Areas with evidence of beaver (Castor canadensis) activity should also be avoided to minimize the threat of chewing through the parachute cord. Bends, sandbars and eddies are sources of non-uniform flow that can result in areas of erosion and /or aggradation and are not ideal deployment locations. Confluences are typically areas of high turbulence and non-uniform flow due to dynamic mixing of water bodies. Due to active mixing, monitoring in confluences is not recommended. Monitor a fair distance downstream to obtain more valid data. Distance downstream will depend on site conditions. Avoid areas of streambank erosion for any stable, robust long-term monitoring stations. Sediment transport can introduce problems related to streambed aggradation and sediment build-up on sensors (Miles 2008; Wagner 2006). Each exact deployment site is GPS

marked and indexed as inconspicuously as possible.

3.2.2 In-situ infrastructure-attached sensor deployment site locations on bridges, aprons, dams / locks and other structures should be deployed at sturdy attachment points. Parameters measured and monitoring objectives should be considered to determine appropriate depths for sensor deployment. For example, conductivity sensors should be deployed as near to the bottom of the river as possible if the goal is to record maximum conductivity readings, whereas sensors should be deployed approximately 1 (one) foot below the surface to record surface temperature. Turbulence and flow disturbance at in stream infrastructure (bridges, etc.) can affect sensor performance and increases maintenance needs. Localized heavy erosion can occur downstream of these structures and aggradation can occur upstream. Avoid deploying sensors at or immediately downstream of outfalls, discharge points and spill-prone areas unless specifically targeting their effects (Miles 2008; Wilde 2006). Each deployment site is GPS marked and indexed as inconspicuously as possible.

3.3 Sensor Maintenance and Deployment Site Visit Intervals

3.3.1 Onset HOBO data loggers

During each deployment site visit the sensor will be retrieved from its PVC sheath and wiped clean using a terry cloth (or similar) rag. At this point data will also be downloaded to a data shuttle and any other required maintenance will be performed (e.g., removal of zebra mussels or algae from sheath; replacement of cable ties, bolts or nuts as needed).

- 3.3.1.1 *In-situ in stream water quality sensors* Deployment site visits will occur no less than every 6 (six) weeks that the sensors are deployed.
- 3.3.1.2 *In-situ infrastructure-attached water quality sensors* Deployment site visits will occur no less than every 60 (sixty) days that the sensors are deployed.
- 3.3.2 YSI EXO2 Datasonde / In-Situ AquaTROLL 500 During each visit the sensor will be retrieved from its PVC sheath and wiped clean using a terry cloth (or similar) rag. Data will be downloaded to a data shuttle and any other required maintenance will be performed (e.g., removal of zebra mussels or algae from sheath).
 - 3.3.2.1 Deployment site visits will occur no less than once every three (3) weeks that the sensors are deployed.

3.4 **Documentation**

- 3.4.1 *Installation Documentation*: Written documentation of all deployment locations records of installation including receipts, owners' manuals, and modifications made, etc. are kept in site files.
- 3.4.2 *Photo documentation*: Complete photographic documentation of each deployment site and installation are recommended. Photos of the site including upstream and downstream and cross-section photos at each sampling site, and photos of installations, etc. should be taken at a variety of site conditions wherever appropriate.
- 3.4.3 *Ongoing Site Visit Documentation:* Records of site visits (in addition to deployment and retrieval information), maintenance performed, problems encountered and their solutions, etc. will be maintained and kept in site files.

4.0 Data Collection, Handling and Management

4.1 Data Collection and Handling

All raw data will be entered into site files by hand or by using sensor specific software packages. Data entry is carried out by the Field Crew Leader.

4.1.1 Sensor Data Retrieval

Data from each sensor will be downloaded to appropriate data shuttles or laptop computers upon sensor retrieval, deployment site visits, or sensor return to ORSANCO offices.

4.1.2 Discrete Data Collection

At the time of sensor deployment and each subsequent deployment site visit (including retrieval) appropriate water quality data will be recorded using a recently calibrated secondary unit (YSI, Hydrolab, In-Situ, etc.) for side by side comparisons with the sensor data to determine accuracy.

4.2 Data Management

All data is housed and backed up on servers located at ORSANCO offices and available for use in relational databases and spreadsheet programs. Findings, data analyses and assessment outcomes (where appropriate) derived from water quality sensor data will be detailed in program-specific reports and made available upon completion and program approval.

5.0 Corrective Action

Immediate Corrective Actions (ICAs) are implemented and resolved expeditiously by leaders in the field. Documentation of ICAs takes place on site and is archived with raw data sheets and on Corrective Action forms when appropriate. In the case of Long Term Corrective Actions (LTCAs), the following steps should be taken:

- 1. Define the problem and discuss with Technical Programs Manager
- 2. Technical Programs Manager assigns responsibility for investigating the problem
- 3. Responsible person(s) determines corrective action to eliminate the problem
- 4. Corrective action plan must be approved by Technical Programs Manager
- 5. Implement corrective action
- 6. Establish effectiveness of corrective action (follow-up)
- 7. Verify that corrective action has eliminated the problem
- 8. Issue report (documentation, CA Form(s)) to Technical Programs Manager

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APPENDIX K

STANDARD OPERATING PROCEDURES

FOR

WATER CHEMISTRY SAMPLE COLLECTION



This document describes the field operations and quality control activities that ORSANCO will use for collecting water chemistry data. It is a step-by-step guide for proper collection, preservation and shipment of river water samples, developed to insure the quality of data collected by field personnel.

1.0 Sampling Locations

ORSANCO will sample at any of 15 randomly selected sites in each of three to four pools as well as fixed sampling locations annually. Sites used in this study are selected following a probability design frame developed for ORSANCO by USEPA.

2.0 Sampling Schedule

Water chemistry will consist of single point samples taken separately from each designated location during the July-October sampling period. Samples are collected so as to arrive at the laboratory on a weekday (Monday through Friday) and within 5 days of collection unless prohibited by laboratory-specified hold times.

4.0 Sample Kit and Field Equipment

The basic field kit required for water chemistry sampling is described below. Replacement supplies are provided by ORSANCO as needed.

TEST EQUIPMENT

In-Situ SmarTROLL handheld sonde or YSI Professional Plus and Pro ODO or comparable multimeter kit

CONTAINERS

2-liter Plastic / Acrylic Kemmerer / Van Dorn sampler with spigot Sample collection containers (provided by laboratory)

MISC SUPPLIES

2 - 50 foot Nylon Ropes Ice Waterproof-Marking Pen Vinyl Tape Zip-Lock Bags Chain of Custody Forms Water Quality Report Forms Safety Glasses Disposable Latex Gloves Strapping Tape Shipping Labels Insulated Coolers USCG Approved PFD Rubber bands

5.0 River Water Sample Collection

- 5.1 *Kemmerer / Van Dorn*: the acrylic sampler will be filled approximately 100 feet from shore and will come from a depth approximately halfway between the surface and bottom. The collection point will be located at the downstream end of the 500m shoreline reach delineating the bounds of the biological collection area. *Surface grab*: surface grab sample containers will be filled approximately 100 feet from shore and will come from a depth approximately 1 foot below the surface at the downstream end of the 500m shoreline reach.
- 5.2 The contract laboratory will provide sample containers labeled with the analytical

parameter, station number, and preservative inside a cooler for shipping. Additionally, each field sampler will mark the bottles with the collection date, time and his/her initials using a waterproof-marking pen.

BOTTLE SIZE	LABEL	PRESERVATIVE
500 mL amber glass	Phenols	H ₂ SO ₄
250 mL plastic	Cyanide (CN ⁻)	NaOH
1 Liter plastic	Direct Ammonia, TOC	H ₂ SO ₄
1 Liter plastic	TSS	None
1 Liter plastic	CI, SO ₄	None
250 mL plastic	Hardness	HNO3

- 5.3 The appropriate amount of river water will be distributed to the sample bottles, capped and inverted several times to thoroughly mix the solution.
- 5.4 Sample bottles will be placed in an insulated cooler with crushed ice to maintain at 4° C.
- 5.5 All pertinent information and field observations will be recorded on Chain of Custody Water Quality Report forms, using one form per station. One copy will accompany the sample to the laboratory and a second copy will be submitted to ORSANCO. Chain of Custody Water Quality Report forms will be sealed in a plastic sealable bag and placed inside the shipping cooler.

6.0 Field Measurements

- 6.1 Temperature, pH, conductivity and dissolved oxygen are measured in the field using portable sondes. Proper calibration and maintenance of these instruments are required to obtain valid data. Instruments will be examined frequently for signs of corrosion or instability and any malfunctions will be reported.
- 6.2 All field measurements, temperature, pH, dissolved oxygen, and conductivity will be made simultaneously with a Multi Probe System/ Data Logger. Units will be calibrated each week of use or more frequently if required following appropriate SOPs.

7.0 Sample Shipment / Delivery

Sample bottles will be shipped or hand delivered to the laboratory in insulated coolers containing sufficient ice to maintain a 4°C temperature during shipment by each sampler. Shipped samples will be sent so that they arrive at the laboratory within 24 hours of collection.

- 7.1 Crushed ice will be obtained locally before beginning sample collections.
- 7.2 As sample bottles are filled, they will be stored in coolers with ice until all locations are sampled. Information will be recorded on the Water Quality Report / Chain of Custody forms.
- 7.3 Place address label and shipping carriers label will be affixed to the cooler lid and secured with clear tape.
- 7.4 Coolers will be delivered to the nearest shipping office for express overnight service to the laboratory.
- 7.5 A copy of the Water Quality Report form / Chain of Custody and a copy of the shipping label will be retained by ORSANCO.

8.0 Quality Control

- 8.1 The interior of all plastic / acrylic samplers (Kemmerer / Van Dorn / surface container) will be kept scrupulously clean. After each sample the container will be emptied, and visually inspected for residual debris or oil films that may contaminate the next sample. If contamination is observed, the sampler will be cleaned with a non-phosphate detergent and thoroughly rinsed with water. Procedures for decontamination will be to clean sampling equipment with a non-phosphate detergent and thoroughly rinse with water. The decontamination procedure will be performed prior to and immediately after sampling and as stated above if contamination is present in the sampling container.
- 8.2 During summer months, additional ice and coolers may be needed to keep samples chilled during shipment or delivery to the laboratory. Fewer bottles will be placed in each cooler and more ice added just before delivery to the carrier. Placing the bottles and ice within a large plastic garbage bag inside the cooler and sealing the bag will help minimize water leakage during shipment.
- 8.3 Containers with chemical preservatives will be handled carefully to prevent spillage on hands and clothing. Acid spills will be neutralized with sodium bicarbonate (baking soda) and the area will be washed with water. Sampling equipment will be cleaned and wiped dry before storage.
- 8.4 Sample blanks and duplicates will be prepared in the field and submitted to the laboratory on a periodic basis. These quality control samples serve as a check on the sampling method, equipment contamination and laboratory performance in meeting analytical precision and accuracy.
- 8.4.1 Sample blanks will be prepared with distilled water following the exact steps used for the routine river samples. Sample bottles will be labeled as "Field Blank" along with the date and time of collection.
- 8.4.2 Duplicates will be prepared by collecting another river water sample and filling a second bottle according to steps outlined under Part 5 above. Sample bottles will be labeled as "Duplicate" along with the date and time of collection.

Parameters	Analytical Method	Detection Limit
Ammonia Nitrogen	350.3	0.03 mg/L
Chloride	325.3	1.0 mg/L
Hardness	SM 2340C	1.0 mg/L
Nitrate + Nitrite	353.3	0.02 mg/L
Phenolics	420.1	0.005 mg/L
Total Kjeldahl Nitrogen	4500-N	0.20 mg/L
Sulfate	HACH 8051	1.0 mg/L
Total Suspended Solids	160.2	1.0 mg/L
Total Phosphorus	365.3	0.01 mg/L
Total Organic Carbon	415.1	0.5 mg/L

Table 4. Analytical Parameters, Methods and Reporting Levels

APPENDIX L

STANDARD OPERATING PROCEDURES

FOR

BOTTOM SEDIMENT SAMPLE COLLECTION

ORSANCO follows EMAP-GRE sediment collecting procedures found in the EMAP-GRE Field Operation Manual, Section 11, p207-214, excerpted in the following document.



At or near each sampling location, a fine-sediment sample is collected using either a hand scoop or a "petite Ponar" grab sampler. The objective is to collect a 4-L composite sample that is representative of MCS depositional areas at the site. The composite sample will be subsampled in the lab for multiple analyses. Section 2.0 describes the sediment sample collection procedures in detail.

1.0 Sample Collection Procedure

- 1.1 Sediment samples are collected at each 100 meter transect within the sampling zone.
- 1.2 Locate sediment samples in areas or patches of fine substrate (silt / sand, silt, clay, muck) in a zone bounded on the shore side by the apparent low-water mark from daily flow fluctuations and bounded on the river side by the 0.3-m (usually about mid-biceps) depth contour (recommended maximum sample depth; deeper sampling may be possible). The low-water mark at a site can often be detected by the presence of periphyton or attached filamentous algae just below the low-water mark. If samples cannot be safely collected by wading at a station due to vertical banks or other reason go to step 2.5.
- 1.3 Be sure to avoid the area that has just been kick sampled. Sampling up-river from kick sample locations is recommended. If fine substrates are not present within 5 m up- or downriver from the station, do not collect sediment at that station and flag the station on the form.
- 1.4 If fine substrate is present, use a small scoop to collect a sample of about 225 cm2 (15 x 15 cm [6 x 6 inches]) of the top 2 cm of substrate (this volume is approximately equal to six scoops). Place the sample in a clean bucket. Use gloves for handling sediment. Do not assume rip rapped shorelines lack fine sediment. Look for fines between the large rocks
- 1.5 If wading is not possible, use a petite Ponar sampler or similar device deployed from the boat to collect a sediment sample adjacent to the station. Release the petite Ponar sample onto a tub and use the scoop to collect about 225 cm2 (15 x 15 cm [6 x 6 inches]) of the top 2 cm of the sample. Estimate sample area visually. Place the subsample in the sediment composite bucket and discard the rest of the Ponar sample.
- 1.6 Repeat steps 2.2-2.5 at each of the 6 littoral stations. Record the total number of replicates (stations) included in the composite. Note in a comment the stations at which sediment was collected using a non-wading method.
- 1.7 It is important that a sufficient sediment (not less than 4 L) sample for analysis be collected. If multiple stations have no fine sediment, it is permissible to collect extra sample at stations that do have fine sediment or between stations. Be sure to note this in a comment.
- 1.8 Using a large stainless steel spoon, thoroughly mix the composite sample in the bucket and transfer 4 L of the composite in a 30 x 50-cm 3-mil thick polyethylene bag. Try to limit the amount of sediment adhering to the inside of the bag near the top. Grasp the bag just above the sediment to express the air. Twist and knot the bag to seal it. Write the site number and date directly on the bag with a permanent marker and place it in a cooler with ice.
- 1.9 Go to section 3 for sample labeling and preservation procedures.

2.0 Sample Labeling and Preservation

2.1 To avoid clutter in the boat, sediment samples may be transported to the ramp or base location (if it is close to the ramp) in a cooler with ice for final labeling and preservation.

2.2 Place the sediment sample inside a second 3-mil polyethylene bag, twist the top, and knot to seal. Prepare a label (Figure 1.) for outside the outer bag. Using a fine-point permanent marker, fill in the site number and sample date. Place the label on the outer bag and cover it with clear tape. Place the sample on ice or in a refrigerator. Do not freeze sediment samples.

SEDIMENT GRAB (SG)
GRW04449
// 200
Composite volume L
Site visit number 1 2
300255

Figure 1. Example sediment sample label.

3.0 QA Considerations

- It is permissible to collect sediment between stations to insure a composite volume of at least 4L. Note deviations from standard procedure in a comment.
- Do not assume rip rapped shorelines lack fine sediment. Look for fines between the large rocks.
- Mix the composite sediment sample thoroughly before extracting the final 4L composite.
- When sampling debris pulled from the river, be sure to sample the upper surface.
- Monitor sediment samples in your possession to insure they do not warm up or freeze.

4.0 Safety Considerations.

- Use extreme care walking on riprap. Rocks can shift unexpectedly and serious falls are possible.
- Use caution when sampling in swift or deep water. Wear a suitable PFD and consider using a safety tether held by an assistant. For most people, conditions are rarely suitable for collecting a periphyton or sediment sample in water deeper than 0.6 m.
- Do not attempt to collect periphyton or sediment from vertical or near vertical banks.
- Professional-quality breathable waders with a belt are recommended for littoral sampling. Neoprene boots are an alternative, but should have sturdy, puncture-resistant soles.
- Use caution using the Ponar-type samplers. The jaws are sharp and may close unexpectedly. Replace frayed lines and worn parts.
- Raise the Ponar sampler from and into a plastic tub rather than from the boat deck. This prevents feet from getting under the sampler.
- Don't try to remove large pieces of debris from the river by yourself.
- Use safety glasses and gloves when handling formalin.
- Avoid contact with sediment samples. Use gloves if necessary.

5.0 References

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