

RECLAMATION

Managing Water in the West

Technical Memorandum No. 86-68220-13-01

Field Protocol: Field Preparation of Water Samples for Dreissenid Veliger Detection

**Field Standard Operating Procedure (Field SOP)
Field SOP Version 4
Date Revised: 2013**

**Bureau of Reclamation
Technical Service Center
Reclamation Detection Laboratory for Invasive
and Native Species**



Raw water sample collected
using a 64 micron plankton tow

MISSION STATEMENTS

The U.S. Department of the Interior protects America's natural resources and heritage, honors our cultures and tribal communities, and supplies the energy to power our future.

The mission of the Bureau of Reclamation is to manage, develop, and protect water and related resources in an environmentally and economically sound manner in the interest of the American public.

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Prepared for:

**Bureau of Reclamation
Technical Service Center
Research and Development Office**

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U.S. Department of the Interior
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REVIEW REQUIREMENT

Part A: Document Does Not Require Review

Explain _____

Part B: Document Requires Peer Review: SCOPE OF PEER REVIEW
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REVIEW CERTIFICATION

Peer Reviewer- I have reviewed the assigned items/section(s) noted for the above document and believe them to be in accordance with the project requirements, standards of the profession, and Reclamation policy.

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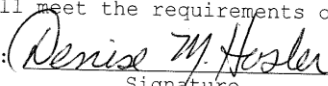
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1. ACRONYMS AND DEFINITIONS

Dreissenid: Genus of freshwater mussel

Zebra Mussel: (*Dreissena polymorpha*) Invasive mussel species

Quagga Mussel: (*Dreissena rostriformis bugensis*) Invasive mussel species

Veliger: For the purpose of this paper, veliger refers to larval zebra-/ quagga mussel

DOI: Department of Interior

Reclamation: Bureau of Reclamation

PFD: Personal Flotation Device

DI water: Deionized water

mL: Milliliter

L: Liter

Plankton tow net: Mesh net used to collect plankton from water samples

Cod end: Weighted end of the plankton tow net

Secchi disk: Circular disk used to measure water transparency

View scope: Plastic tube used to assist in Secchi disk reading

YSI: Multi-probe used to collect water quality data

Handheld: The computer unit of the YSI

Sonde: YSI submerged unit, containing probes

Sonde cup: Storage and calibration cup for the sonde

Sonde cage: Protection cage for the sonde

DO probe: Dissolved oxygen probe on YSI sonde

2. SCOPE AND APPLICABILITY

This standard operating procedure (SOP) is used to establish a uniform format for duties performed by the zebra/ quagga mussel field sampling crew. This method is applicable to collection of YSI data and dreissenid water samples that are shipped to the mussel lab at the Bureau of Reclamation Technical Service Center, Denver CO. The goal of this SOP is to standardize how each task in the field is performed by every crew member. This SOP is not a replacement for training.

3. SUMMARY OF METHOD

Collection of water quality data using the YSI and collection and shipment of dreissenid water samples.

4. INTERFERENCES

The main interference in early mussel detection is cross contamination from other samples and water bodies. In order to mitigate this interference, cleaned, sanitized and well-labeled bottles will be used for collection. To ensure that no mussel larvae remain in the net between sites, always soak the plankton tow net and cod end in a vinegar bath between sample sites. Always wash net and cod end with vinegar and bleach at the end of the sampling day and at the end of the sampling trip. Always keep nets for waters contaminated with veligers separate from other nets. A secondary interference in early mussel detection is improper sample handling in the field resulting in lag times between sample collection and shipping. Improper sample handling also can result in a low pH which will hinder detection. When hauling a boat to different reservoirs, make sure that boats are traveling only between waters with no mussel larvae identified. All boats traveling from one negative water body to another negative water body

must be cleaned, drained, and dried as per the Colorado Parks and Wildlife (2012). It is mandatory that all boat handlers learn how to decontaminate boats that may be infected with adult mussels or mussel larvae (appendix A). Refer to the particular states ANS plan for state specific decontamination requirements.

5. CHEMICAL HAZARDS

- 5.1. Distilled white vinegar: CAS No-8028-52-2 UN No: Not Regulated Hazard Class: Not Regulated
 - 5.1.1. Handling: None specified by manufacturer, if spilled, water may be used to dilute
 - 5.1.2. Storage: None specified by manufacturer
 - 5.1.3. Engineering Controls: None specified by manufacturer
 - 5.1.4. First Aid: None specified by manufacturer. Inhalation: Remove to fresh air. Support breathing if needed. Skin: Flush with copious amounts of water, call physician. Eye: Flush immediately with water for 15 to 20 minutes. Call a physician. Ingestion: Large amounts of water should be consumed to dilute. Do not induce vomiting. Do not give emetics or baking soda. Call a physician (MSDS.79839. 2011)
- 5.2. Clorox Regular Bleach: EPA Reg. No. 5813-50
 - 5.2.1. Health Hazard Data: Corrosive: May cause irritation or damage to eyes and skin. Vapor or mist may irritate. Harmful if swallowed. No special protection or precautions have been identified with using this product under directed consumer use conditions
 - 5.2.2. First Aid: Eyes: Hold eye open and rinse with water for 15 to 20 minutes. Remove contact lenses, after first five minutes. Continue rinsing eye. Call physician. Skin: Wash skin with water for 15 to 20 minutes. If irritation develops, call a physician. Ingestion: Do not induce vomiting. Drink a glassful of water. If irritation develops, call a physician. Do not give anything by mouth to an unconscious person. Inhalation: Remove to fresh air. If breathing is affected, call a physician.
 - 5.2.3. Spill Procedures: Control spill. Use absorbents to clean spill. Wash area and let dry. Dispose of in accordance with all applicable federal, state and local regulations. (MSDS EPA Reg. No. 5813-50. 08/09).
- 5.3. Ethanol, Denatured with up to five percent V.V Ether: CAS-No-60-29-7 UN-No1170 Hazard Class: 3
 - 5.3.1. Handling: Wash hands thoroughly after handling. Use only in a well-ventilated area. Take precaution to avoid static discharges. Avoid contact with eyes, skin, and clothing. Do not breathe fumes. Empty containers retain product residue (liquid and/or vapor), and can be dangerous. Keep container tightly closed. Avoid contact with heat, sparks and flame. Avoid ingestion and inhalation. Do not pressurize, cut, weld, braze, solder, drill, grind, or expose empty containers to heat, sparks or open flames.
 - 5.3.2. Storage: Keep away from heat, sparks, and flame. Keep away from sources of ignition. Store in a tightly closed container. Keep from contact with oxidizing materials. Store in a cool, dry, well-ventilated area away from incompatible substances. Store in a designated area marked flammables. Do not store near perchlorates, peroxides, chromic acid or nitric acid.
 - 5.3.3. Engineering Controls: Use explosion-proof ventilation equipment. Facilities storing or utilizing this material should be equipped with an eyewash facility and a safety shower close to workstation location. Use adequate general or local exhaust ventilation to keep airborne concentrations below the permissible exposure limits.

- 5.3.4. Personal Protective Equipment: Eyes: Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166. Skin: Wear appropriate protective gloves to prevent skin exposure. Clothing: Wear appropriate protective clothing to prevent skin exposure. Respirators: A respiratory protection program that meets OSHA's 29 CFR 1910.134 and ANSI Z88.2 requirements or European Standard EN 149 must be followed whenever workplace conditions warrant a respirator's use (MSDS, No. 270660000, 2010).
- 5.4. Sodium Bicarbonate: CAS-No 144-55-8, UN-No Not Regulated, Hazard Class-Not Regulated
- 5.4.1. Handling: Use with adequate ventilation. Minimize dust generation and accumulation. Do not get in eyes or skin. Do not ingest or inhale.
- 5.4.2. Storage: Store in cool, dry place
- 5.4.3. Engineering Controls: Facilities storing or utilizing this material should be equipped with an eyewash facility and a safety shower close to workstation location. Use adequate ventilation to keep airborne concentration low.
- 5.4.4. Personal Protective Equipment: Eyes: Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166. Skin and clothes: Wear appropriate protective clothing to prevent skin exposure. Respirators: A respiratory protection program that meets OSHA's 29 CFR 1910.134 and ANSI Z88.2 requirements or European Standard EN 149 must be followed whenever workplace conditions warrant a respirator's use (MSDS. No 20970, 2008).
- 5.5. Buffer Solutions: pH 4, pH 7, pH 10, UN-No Not Regulated, Hazard Class-Not Regulated
- 5.5.1. Handling: Wear personal protective equipment. Ensure adequate ventilation. Do not breathe vapors or spray mist. Avoid contact with skin, eyes and clothing.
- 5.5.2. Storage: Keep containers tightly closed in a dry, cool and well-ventilated place.
- 5.5.3. Engineering Controls: Ensure adequate ventilation, especially in confined areas. Ensure that eyewash stations and safety showers are close to the workstation location.
- 5.5.4. Personal Protective Equipment: Eyes: Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166. Skin/Clothing: Wear appropriate protective gloves and clothing to prevent skin exposure. Respirators: Follow the OSHA respirator regulations found in 29 CFR 1910.134 or European Standard EN 149. Use a NIOSH/MSHA or European Standard EN 149 approved respirator if exposure limits are exceeded or if irritation or other symptoms are experienced (MSDS. No C2948. 2006).
- 5.6. Conductivity Standard: UN-No Not Regulated, Hazard Class-Not Regulated
- 5.6.1. Handling: Ensure adequate ventilation. Do not breathe vapors or spray mist. Avoid contact with skin, eyes and clothing.
- 5.6.2. Storage: Keep containers tightly closed in a dry, cool and well-ventilated place.
- 5.6.3. Engineering Controls: Ensure adequate ventilation, especially in confined area. Ensure that eyewash station and safety showers are close to the workstation location.

- 5.6.4. Personal Protective Equipment: Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166. Skin/Clothing: Wear appropriate protective gloves and clothing to prevent skin exposure. Respirators: Follow the OSHA respirator regulations found in 29 CFR 1910.134 or European Standard EN 149. Use a NIOSH/MSHA or European Standard EN 149 approved respirator if exposure limits are exceeded or if irritation or other symptoms are experienced (MSDS. No 09-328-2.2010).

6. ENVIRONMENTAL HAZARDS

Outside temperatures can range from 30°F to 120°F, with water temperatures as low as 40°F during the field season. Be aware of predicted weather conditions; avoid water travel if conditions are too extreme. There is little to no shelter on most of the boats, so individuals will need to minimize sun exposure by wearing the appropriate attire. Sun screen, sunglasses, hat with brim, long sleeve shirt, long pants, close toed shoes, etc. Protective sun wear will be the responsibility of each person. Sun screen, insect repellent, gloves and drinking water will be provided by the field crew leader.

- 6.1. Bad weather: rain, high winds, rough water, cold or hot temperatures
 - 6.1.1. Dress appropriately, and in layers so that a comfort level can be obtained
 - 6.1.2. Wear insulated clothing or rain gear in colder weather
 - 6.1.3. Wear summer appropriate attire
 - 6.1.4. Avoid cotton clothing that might absorb water and wick body heat away
 - 6.1.5. Wear footwear with non-skid soles to reduce slips and falls
- 6.2. Heat exposure and dehydration
 - 6.2.1. Drink enough water
 - 6.2.1.1. One quart every two hours minimum
 - 6.2.2. Reduce heat exposure by covering the areas exposed with a hat and sleeved-shirts
- 6.3. Sunburn (UV)
 - 6.3.1. Wear sunscreen of SPF 15 or greater
 - 6.3.2. Wear a hat with a brim to reduce heat exposure on face
 - 6.3.3. Wear a long sleeve shirt to reduce sun exposure to skin
- 6.4. Bright sun (glare)
 - 6.4.1. Wear sunscreen of SPF 15 or greater
 - 6.4.2. Wear a hat with a brim to reduce heat exposure on face and glare
 - 6.4.3. Wear sunglasses
- 6.5. Poisonous animals: Snakes, arachnids, and other arthropods
 - 6.5.1. If needed, wear insect repellent
 - 6.5.2. Wear close toed shoes
 - 6.5.3. Notify field crew leader if any poisonous animals are in the area
 - 6.5.4. Get immediate medical attention if bitten by a snake or if any rashes or irritation occurs (Field JHA Template 2010).

7. FIELD HAZARDS

7.1. Minor injuries

7.1.1. For minor cuts and scrapes, a first-aid kit is located on the boat and in government vehicles

7.2. Serious injuries

7.2.1. Notify the local state authorities immediately of any serious incident

7.2.2. If needed, use 911 services

7.3. Heavy lifting: Ice chests, tool boxes, YSI equipment, sampling equipment

7.3.1. Any equipment weighing more than 50 lbs. should be lifted by 2 people

7.3.2. Always lift using the knees and not the back

7.3.3. Use caution when loading and unloading equipment to and from the boats. Use two people to carry equipment when possible (Field JHA Template 2010).

8. VEHICLE HAZARDS

Bureau of Reclamation (Reclamation) vehicles will only be operated by personnel who have current driver training provided by Department of Interior (DOI).

8.1. Shifting loads

8.1.1. Make sure loads are tied down properly

8.2. Towing

8.2.1. Boat towing will only be performed by individuals who have passed the Department of Interior's Motorboat Operator Certification Course (MOCC).

8.2.2. Make sure vehicle is rated to haul a boat

8.2.3. Be aware that hauling affects vehicle performance

8.2.4. Ensure all brake lights, surge brakes and turn signals are in working order

8.3. Long driving hours

8.3.1. If tired, pull over

8.3.2. Drink plenty of water

8.3.3. Keep ambient temperature of the vehicle cool and comfortable. Too hot or too cold interiors can make the driver tired and distracted (Field JHA Template 2010).

9. BOAT HAZARDS

Reclamation boats will only be operated by personnel who have current MOCC training provided by the DOI.

9.1. Heat stress

- 9.1.1. Drink plenty of water
- 9.1.2. Eat a snack
- 9.1.3. Wear sunscreen (SPF 15 or greater)
- 9.1.4. Wear sunglasses
- 9.1.5. Wear appropriate attire to reduce sun exposure

9.2. Cold, hypothermia

- 9.2.1. Wear warm clothing in layers
- 9.2.2. Wear materials that don't absorb water and wick away heat from the body
- 9.2.3. Wear gloves to reduce water contact
- 9.2.4. Wear warm non-skid soled shoes

9.3. Falling overboard

- 9.3.1. In waters over three feet deep a personal floatation device (PFD) is required
 - 9.3.1.1. The PFD will be obtained from the field crew leader or will be provided on the boat
- 9.3.2. Note the swimming ability of every person and also their comfort level on the boat prior to launching
- 9.3.3. Make sure all coast guard required equipment is on the boat
 - 9.3.3.1. Fire extinguisher
 - 9.3.3.2. Throwable PFD (Type IV)
 - 9.3.3.3. Bell, whistle, horn
 - 9.3.3.4. Visual distress signal (flares)
 - 9.3.3.5. Paddle or oar
 - 9.3.3.6. Marine radios, Channels 16 and 22 are for emergencies (Field JHA Template, 2010).

10. TRAINING

- 10.1. Boat training and orientation will be provided on site as needed
- 10.2. Watercraft inspection and decontamination training is available and advisable for sampling in positive water bodies. See the state ANS plan for further details
- 10.3. Reclamation vehicles will only be operated by personnel who have current driving training
- 10.4. Boat towing and operation will only be operated by personnel who have successfully completed the Department of the Interior's MOCC training
- 10.5. Sample collection training will be provided to personnel on site as needed (Field JHA Template 2010).

11. MATERIALS

11.1. Boating

- 11.1.1. Personnel floatation devices (PFD)
- 11.1.2. Fire extinguisher
- 11.1.3. Throwable PFD (Type IV)
- 11.1.4. Bell, whistle, horn
- 11.1.5. Visual distress signal (flares)
- 11.1.6. Paddle or oar
- 11.1.7. Proper attire: Including sunglasses, hat with brim, sunscreen, long sleeved shirt and long pants
- 11.1.8. Marine radio or cell phone

11.2. Sampling

- 11.2.1. Plankton net (64- μ m mesh) with weighted COD-end piece (64- μ m mesh)
 - 11.2.1.1. Reel of rope, marked in 1 meter increments up to 50 meters
- 11.2.2. Sample bottles (appendix B)
 - 11.2.2.1. Sterilized, clean and pre-labeled
- 11.2.3. Secchi disk
 - 11.2.3.1. Reel of rope marked in 1/10 meter increments
 - 11.2.3.2. View scope
- 11.2.4. Multiprobe water quality instrument
 - 11.2.4.1. YSI sonde
 - 11.2.4.2. YSI handheld
 - 11.2.4.3. YSI cord
 - 11.2.4.4. YSI cage
- 11.2.5. Bucket for easy cord handling
- 11.2.6. Anemometer (if applicable)
- 11.2.7. Permanent marker
- 11.2.8. Deionized water (DI water)
- 11.2.9. Bleach
- 11.2.10. Vinegar
- 11.2.11. Decontamination buckets for plankton tow nets
- 11.2.12. Baking soda (Sodium Bicarbonate) for buffer
- 11.2.13. Plastic, 0.1 gram, spoons to measure buffer
- 11.2.14. Field notebook
- 11.2.15. Pencils/ pens
- 11.2.16. Paper towels
- 11.2.17. Alcohol to preserve samples
- 11.2.18. Tape measure

11.3. Shipping

- 11.3.1. Sealable plastic bags
 - 11.3.1.1. Quart and gallon bags
- 11.3.2. Garbage bags
- 11.3.3. Diapers
- 11.3.4. Ice cooler

- 11.3.5. Electrical tape to seal sample bottles
- 11.3.6. Packing tape to seal coolers
- 11.3.7. FedEx shipping form (appendix C)

12. REAGENTS AND CHEMICALS

- 12.1. Concentrated alcohol
- 12.2. Lake water preserved with alcohol
 - 12.2.1. Twenty percent alcohol added in field
- 12.3. Vinegar
 - 12.3.1. Use to decontaminate the plankton tow net
- 12.4. Bleach
 - 12.4.1. Use to decontaminate the plankton tow net
- 12.5. Buffers
 - 12.5.1. Sodium bicarbonate (baking soda)
 - 12.5.2. Calibration buffers for pH (7 and 10)
 - 12.5.3. Calibration standard for conductivity

13. FIELD SAMPLE PREPARATION

- 13.1. Label bottles (appendix B and D)
 - 13.1.1. State, water body, sample site, depth, # of tows, date
- 13.2. Label sample bottle lids
 - 13.2.1. State, water body, sample site
- 13.3. Inspect plankton tow net
 - 13.3.1. Look for holes, rips or tears
 - 13.3.2. Check metal/ PVC collar to make sure cod end screws on securely
 - 13.3.3. Make sure the mesh is fitted to the inside of cod end
 - 13.3.4. Make sure all the knots are securely tightened
- 13.4. Calibrate YSI each morning (appendix D and E)
 - 13.4.1. Calibrate conductivity
 - 13.4.2. Calibrate pH (7 and 10)
 - 13.4.3. Calibrate dissolved oxygen on site of each reservoir
 - 13.4.4. Calibrate the pressure/ depth on site of each reservoir

14. SAMPLING

14.1. Secchi Disk

- 14.1.1. Lower Secchi disk with a line marked at 1/10 meter increments
- 14.1.2. Lower the disk until the black and white markings are undistinguishable
- 14.1.3. If the Secchi disk depth exceeds one meter, use the view scope to look at the disk
- 14.1.4. Record the depth with and without the view scope in the field notebook

14.2. YSI Sampling (appendix D and E)

- 14.2.1. Calibrate YSI every morning
- 14.2.2. Calibrate DO and pressure/ depth on site
- 14.2.3. Turn on handheld by pressing the green power button
 - 14.2.3.1. 650 Main Menu. Select 'Sonde Run'. Enter
 - 14.2.3.2. 'Log one sample'. Enter
 - 14.2.3.3. Pick site. Enter. Pick filename
 - 14.2.3.4. 'Sample Logged' will blink multiple times
 - 14.2.3.4.1. Try to keep cord stationary during this time
 - 14.2.3.5. 'Sample Logged' will be replaced with '650'
 - 14.2.3.6. The YSI cord can be lowered one meter
- 14.2.4. Repeat steps 14.2.3.2 to 14.2.3.6 in one meter increments until the bottom is reached
- 14.2.5. Record the maximum depth in the field notebook
- 14.2.6. At the end of each day/ trip, upload the data from the YSI hand held to a laptop or computer (appendix F)

14.3. Plankton Tow Net Sampling

- 14.3.1. Make sure the sample bottles and lids are pre-labeled (section 13.1 to 13.3, appendix B and D)
- 14.3.2. Keep plankton tow net in a bucket with vinegar for decontamination
 - 14.3.2.1. Dip net and cod end in water before sampling next site to rinse off vinegar
- 14.3.3. Reference the total depth from the YSI readings. Round down to the nearest whole number
 - 14.3.3.1. Example: If the total depth of the reservoir is 13.784 meters, lower net to 13 meters
- 14.3.4. Record total depth in the field notebook
- 14.3.5. Let the net drop down to the depth in 14.3.3
- 14.3.6. Pull the plankton tow net slowly (hand-over-hand) back up to the surface
 - 14.3.6.1. If the cod end is filled with sediment, re-sample and raise the net by half a meter
- 14.3.7. Dunk the plankton net two or three times without submerging the top ring to rinse the contents from the net into the cod end
- 14.3.8. Unscrew the cod end, gently swirl the sample around to let some of the water drain out, and pour the water into the appropriate pre-labeled sample bottle
- 14.3.9. Using a squirt bottle with DI water, rinse the cod end and pour the wash water into the sample bottle, three times
 - 14.3.9.1. If there is too much liquid in the bottle, re-pour into the cod end to drain some fluid
- 14.3.10. Repeat 14.3.5 to 14.3.8, five times
- 14.3.11. Allow one to two inches of empty space in the sample bottle for preservation

- 14.3.12. Add 0.2 grams or 0.2mLs of baking soda per 100 mL of liquid in the sample bottle
 - 14.3.12.1 If using a plastic 0.1 mL measuring spoon, add two 0.1 mL spoonfuls per 100 mLs.
 - 14.3.12.2 Gently shake the sample to mix the buffer
- 14.3.13. Add 20 percent of the sample volume of alcohol in order to preserve the sample
 - 14.3.13.1. Measure the height of the liquid with tape measure. Multiply the height, in inches, by 0.2
 - 14.3.13.2. The result of this equation is the amount of alcohol, in inches, that should be added to the sample
- 14.3.14. Gently shake the sample to mix the preservative and buffer
- 14.3.15. Release built up pressure in the bottle by opening the lid of the bottle
- 14.3.16. Place sample in cooler with ice
- 14.3.17. Decontaminate the plankton tow net in a bucket with vinegar between sites
- 14.3.18. Remove residual vinegar by rinsing the net in lake water before sampling
- 14.4. **Algal Toxin Sampling** (appendix G)
 - 14.4.1. Sampling done on request

15. END OF DAY - TRIP PROCEDURES

- 15.1. When traveling between negative water bodies: clean, drain and dry the boat
 - 15.1.1. Training on watercraft inspection and decontamination is available and advisable when sampling in positive water bodies
 - 15.1.2. See the state ANS plan for the states requirements for decontaminating boats
- 15.2. Remove plankton tow net from the vinegar bath and rinse well with water
- 15.3. Quick rinse with bleach and rinse well with water
 - 15.3.1. Leaving net in bleach too long will cause the net to tear
- 15.4. Rinse off Secchi disk
- 15.5. Rinse off sonde cage and probes
 - 15.5.1. Place wet sponge and black cap over DO probe
 - 15.5.2. Fill sonde cup to line
 - 15.5.3. Place sonde cup onto sonde
 - 15.5.4. Place water samples on ice in the cooler
 - 15.5.4.1. Or keep samples in the hotel mini-fridge

16. NET DECONTAMINATION (appendix D)

- 16.1. Between sites on the reservoir
 - 16.1.1. Soak net and cod end in a vinegar bath between sample sites
 - 16.1.2. Rinse with lake water before sampling to remove vinegar
- 16.2. Between reservoirs
 - 16.2.1. Wash net and cod end in vinegar and rinse with water
 - 16.2.2. Spray net with bleach solution and rinse with water
 - 16.2.2.1. Leaving net in bleach too long will cause the net to tear
- 16.3. End of sampling trip
 - 16.3.1. Wash net and cod end in bleach
 - 16.3.2. Rinse with water to remove bleach
- 16.4. Use dedicated nets for unconnected water bodies
- 16.5. Use dedicated nets for positive waters

17. SHIPMENT

- 17.1. Cooler preparation
 - 17.1.1. Make sure that drain valve on cooler is taped closed
 - 17.1.2. Make sure cooler is clean and dry
 - 17.1.3. Open a trash bag and lay it in the cooler to contain any leaks that may occur
 - 17.1.4. Make sure ice or blue ice is packed to keep sample cool during shipping
- 17.2. Sample bottle preparation
 - 17.2.1. Make sure that all labels are easily legible
 - 17.2.2. Make sure the lid has the state and site location written on it
 - 17.2.3. Make sure that site location, sample date and tow length and number of tows are recorded on the sample bottle (appendix B)
 - 17.2.4. Wrap electrical tape once around the lid of the bottle to prevent it from leaking
 - 17.2.5. Place diaper over sample bottle
 - 17.2.6. Place sample in garbage bag lined cooler with enough ice and / or blue ice to keep the samples chilled
- 17.3. Shipping preparation
 - 17.3.1. Once all samples are inside, close garbage bag tightly, and tie a knot to close the bag to prevent spills during shipping
 - 17.3.2. Tape lid shut
 - 17.3.3. Tape sides of cooler to help contain leaks
 - 17.3.4. Check the drain valve and make sure it is taped closed
 - 17.3.5. Make sure the FedEx form is properly filled out (appendix C)
 - 17.3.6. Overnight the cooler to the Reclamation lab for analysis
 - 17.3.6.1. Notify lab manager if shipping on a Friday

18. PERMITS

18.1. A scientific sampling permit must be renewed each year and carried at all times while conducting sampling

18.1.1. Ask lab manager if a permit is required before sampling

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20. CONTACTS

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Appendix A- Boat Decontamination

Ask the state ANS coordinator the decontamination procedures for that state. The following are guidelines on clean, drain and dry as well as hot pressure washing boats. All field crews leaders are boater certified and have received training on how to decontaminate boats through the DOI. Further information and certification can be located at www.100thmeridian.org. Get full boat decontamination procedures from the state ANS coordinator.

Boats that travel between negative water bodies need to follow the clean, drain and dry procedures. The following are short directions on how to clean, drain and dry a boat (Colorado Parks and Wildlife 2012).

1. **Clean**
 - 1.1. Remove any plant, plant fragments, mud or other debris
 - 1.2. Check trailer, axels and wheels in and around the boat itself
 - 1.3. Check anchor, props, jet engines, ropes, bumpers, paddles
 - 1.4. Clean, check and dry all parts and equipment that came in contact with water
 - 1.5. Empty bait buckets in trash
2. **Drain**
 - 2.1. Drain every space or item that can hold water
 - 2.2. Follow factory guidelines for removing water from engines
 - 2.3. Remove the drain plug and drain the water out
 - 2.4. Drain live-wells, bilge, ballast tanks and transom wells
3. **Dry**
 - 3.1. Allow everything to dry completely before launching into another negative water body

Boats that have contact with waters that are positive or suspect, must be quarantined for at least 30 days. The following are short directions on how to decontaminate a boat using a hot pressure washer.

1. Find a location where the waste water can be contained and not re-enter the water system in any way.
2. Use a presser washer with water at a temperature of greater than 140°F
 - 2.1. Water loses 15-20°F per foot of distance when sprayed, adjust accordingly
 - 2.2. Power washer needs to spray at least 4 gal/min
 - 2.3. Nozzle pressure should be greater than 3000 psi, but not exceed 3500 psi
3. Concentrate spray stream for at least 60 seconds
 - 3.1. Use the flushing attachment for the live well, bait well, storage compartments, bilge area, ballast tanks, bladders, gear and equipment
 - 3.2. Use a brush to wash hard to reach places
4. To clean engine
 - 4.1. Start engine, idle for one to two minutes while running pressure washer in engine compartment
 - 4.2. Beware of overheating. Only let engine run one to two minutes at a time
 - 4.3. Keep an eye on engine temperature to keep from overheating (Zook and Phillips 2009)

Appendix B- Bottle Labels

State
Water body
Site Location
Sample Date
0-depth (meters) x # of tows

Sample Bottle Lid Label

State
Water body
Site Location

Appendix C- Fed Ex Shipping Form

<p>From: Date: _____ FedEX Account Number _____ Name: _____ Phone Number _____</p> <p>Company: <u>BOR 84-21310</u></p> <p>Address: <u>6th and Kipling Bldg 67 Rm 152</u> City: <u>Lakewood</u> State: <u>CO</u> Zip: <u>80228</u></p> <p>To: Recipients Name: _____ Date: _____ Company: <u>BOR</u> Address: <u>6th and Kipling Bldg 56 Rm 2010 86-68222</u> City: <u>Denver</u> State: <u>CO</u> Zip: <u>80225</u></p>	<p>Package Service</p> <p><input checked="" type="checkbox"/> Overnight</p>
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Appendix D- Reference Notecards

May be fixed to notecard, laminated and taken into the field.

Calibrate YSI	
Calibrations to do every morning:	Calibrations to do at each reservoir:
1. Calibrate Conductivity	1. Calibrate DO. Allow 5-10 min for calibration to finish
2a. Calibrate pH (7.0) 2b. Calibrate pH (10.0)	2. Calibrate Pressure. Set depth for 0.1 meters
Edit Site List	Out of Range- Accept Anyway?
a. 650 Main Menu b. Select 'File' c. Select 'Logging Setup' d. Select 'Edit Site List'	a. Always select 'No' b. Make sure cup is secure c. Re-try the calibration d. Select 'Clean Optics' in DO calibration menu

Net Decontamination
Between Sample Sites 1. Soak net and cod end in vinegar bath between sample sites 2. Rinse with water before sampling to remove vinegar
Between Reservoirs 1. Wash net and cod end in vinegar and rinse with water 2. Spray net and cod end with bleach and rinse with water <ul style="list-style-type: none"> • Prolonged exposure to bleach will shorten life of net
End of Sampling Trip 1. Wash net and cod end in vinegar and rinse with water 2. Wash net and cod end with bleach and rinse with water
Dedicate nets for unconnected water bodies
Dedicate nets for positive waters

Field Sample Preparation

1. Label Bottle:

State, Water body, Sample Site, Depth, Number of tows, Date

2. Label Sample Bottle Lid:

State, Water body, Sample Site

4. Inspect Plankton tow net

5. Calibrate YSI

Sampling Procedures

YSI Sampling	Plankton Tow Net Sampling	Secchi Disk
Calibrate DO and Pressure on site	Keep net and cod end in vinegar	Lower Secchi disk with a line marked in 1/10 meter increments
Select 'Sonde Run' on Handheld	Dip net and cod end in water to rinse off vinegar	Lower the disk until the black and white markings are no longer visible
Select 'Log one Sample'	Screw cod end onto net	If the disk exceeds 1 meter, use the view scope to increase visibility
Pick site. Pick File name	Reference YSI total depth and lower net down to nearest meter	Record the depth with and without the view scope in the field notebook
'Sample Logged' will blink	Pull net up slowly hand over hand	
When '650' replaces 'Sample Logged' in screen	Repeat for a total of 5 pulls	
Lower YSI 1 meter	Add 20% alcohol to preserve sample	
	Add 0.2 grams baking soda per 100 ml of sample	

Appendix E- Calibrate YSI

1. Calibrate the YSI every morning. Calibrate pressure/ depth and DO probes on location
 - 1.1. Connect the handheld to the YSI cord
 - 1.2. Attach the sonde to the other end of the YSI cord
 - 1.2.1. Make sure attachments are secure
 - 1.3. Turn on handheld by pressing the green power button
2. Calibrate conductivity
 - 2.1. Remove sonde cup. Remove black cap and sponge from DO probe (the large probe)
 - 2.2. Rinse sonde cup with tap water
 - 2.2.1. Blot DO probe with a paper towel
 - 2.3. Rinse cup with a small amount of conductivity standard
 - 2.3.1. Dispose down drain
 - 2.4. Fill cup half full of conductivity standard
 - 2.5. Replace cup on sonde, making sure cup is tight. If it is not on tight, it could alter the readings
 - 2.6. On the handheld
 - 2.6.1. '650 Main Menu'
 - 2.6.2. Select 'Sonde Menu'. Enter
 - 2.6.3. Select 'Calibrate'. Enter
 - 2.6.4. Select 'Conductivity'. Enter
 - 2.6.5. Select 'Sp Cond'. Enter
 - 2.6.6. If needed, enter conductivity value, found on bottle. Enter
 - 2.6.6.1. Check if a conversion is needed
 - 2.6.7. Select 'Calibrate'. Enter
 - 2.7. Handheld will display the word 'Calibrated' in upper right corner when finished
 - 2.8. Escape back to 'Calibrate' menu
 - 2.8.1. Replace conductivity standard in bottle
 - 2.8.2. Rinse cup and probes with tap water, dispose down drain
 - 2.8.3. Blot DO probe with paper towel
3. Calibrate pH (7.0 pH)
 - 3.1. Rinse cup with a small amount of 7.0 pH standard
 - 3.1.1. Dispose down drain
 - 3.2. Fill the sonde cup about half full of 7.0 pH standard
 - 3.3. Replace cup on sonde making sure cup is on tight. If it is not tight, it could alter the readings
 - 3.4. On handheld, select 'ISEI pH'. Enter
 - 3.4.1. Select '2 point'. Enter
 - 3.4.2. 1st pH, type '7.0'. Enter
 - 3.4.3. Select 'Calibrate'. Enter
 - 3.4.4. Handheld will display the word 'Calibrated' in the upper right corner when finished
 - 3.4.5. Select 'Continue'. Enter
 - 3.4.6. Replace 7.0 pH standard in bottle
 - 3.4.7. Rinse sonde cup and probes with tap water, dump rinse water down the drain
 - 3.4.8. Blot DO probe with paper towel
4. Calibrate pH (10.0 pH)
 - 4.1. Rinse cup with a small amount of 10.0 pH standard
 - 4.1.1. Dispose down drain
 - 4.2. Fill the sonde cup about half full of 10.0 pH standard

- 4.3. Replace cup on sonde making sure cup is on tight. If it is not on tight, it could alter the readings
- 4.4. On handheld
 - 4.4.1. 2nd pH, type '10.0'. Enter
 - 4.4.2. Select 'Calibrate'. Enter
 - 4.4.3. Handheld will display the word 'Calibrated' in the upper right corner when finished
 - 4.4.4. Continue. Enter
 - 4.4.5. Replace 10.0 pH standard in bottle
 - 4.4.6. Rinse sonde cup and probes with tap water, dump rinse water down the drain
 - 4.4.7. Blot DO probe with paper towel
- 4.5. Replace sponge and black cap on the DO probe
- 4.6. Fill cup with a small amount of tap water, enough to cover the bottom of the cup, but not enough to touch the DO probe
5. Calibrate DO
 - 5.1. Dissolved oxygen (DO) measurements require a barometric reading. Therefore, do this calibration on the first site of every water body
 - 5.2. At Calibration menu, select 'Optic C-Dissolved Oxygen'. Enter
 - 5.2.1. Select 'ODO Sat%'. Enter
 - 5.2.2. When handheld shows the barometric pressure. Enter
 - 5.2.3. Fill cup with a small amount of tap water, enough to cover the bottom of the cup, but not enough to touch the DO probe
 - 5.2.4. Replace sonde cup, do not screw on tightly
 - 5.2.5. Select 'Calibrate'. Enter
 - 5.2.6. Allow the unit to sit for 5-10 minutes to allow DO probe to measure the vapor pressure
6. Calibrate pressure and depth
 - 6.1. Calibrate these measurements at the first site of every water body
 - 6.2. At Calibration menu, select 'Pressure'
 - 6.2.1. Replace the sonde cup with the sonde cage and lower the YSI until the water is covering the cage.
 - 6.2.2. Calibrate the depth to 0.1 meters. Enter
 - 6.3. Escape back to '650 Main Menu'. YSI is now ready to run
7. Out of range-accept anyway?
 - 7.1.1. Always select 'No'
 - 7.1.2. Make sure cup is secure
 - 7.1.3. Make sure standards are touching the probes
 - 7.1.4. Make sure units for calibration are correct on the handheld
 - 7.1.5. Make sure the proper standard is being used
 - 7.1.6. Re-try the calibration
 - 7.1.7. Select 'Clean optics' in the DO calibration menu
 - 7.1.8. Use different standards (if available)
8. Edit site list
 - 8.1. 650 Main menu on handheld
 - 8.1.1. Select 'File'. Enter
 - 8.1.2. Select 'Logging Setup'. Enter
 - 8.1.3. Select 'Edit Site List'. Enter
 - 8.1.4. Add, change or delete sites here

9. Run a sample
 - 9.1. Turn on handheld by pressing the green power button
 - 9.2. 650 Main Menu. Select 'Sonde Run'. Enter
 - 9.2.1. 'Log one sample'. Enter
 - 9.2.2. Pick site. Enter. Pick filename (section 14.2 YSI sampling and appendix D section 8)
 - 9.2.3. 'Sample Logged' will blink multiple times
 - 9.2.4. Try to keep cord stationary during this time
 - 9.2.5. 'Sample Logged' will be replaced with '650'
 - 9.2.6. The YSI cord can be lowered one meter
 - 9.3. Repeat steps 9.2.1 to 9.2.7 in one meter increments until the bottom is reached
 - 9.4. Record the maximum depth in the field notebook
 - 9.5. At the end of each day/trip, upload the data from the YSI hand held to a laptop or computer
10. How to view files
 - 10.1. '650 Main Menu'. 'File'. Enter
 - 10.2. 'View File'. Enter
 - 10.3. Pick file name to view. Enter
 - 10.4. View YSI data for that file name

Appendix F- Export data from handheld to Excel

1. Open EcoWatch program on laptop
2. Plug YSI handheld into laptop
 - 2.1. Turn on handheld by pressing green power button
3. In EcoWatch, click on the dynamite icon
4. Select the correct COM port
 - 4.1. A new window will open with a '#' sign on the page
5. On the handheld go to the 'Main Menu' screen
 - 5.1. Select 'File'
 - 5.2. Select 'Upload to PC'
 - 5.3. Select file name
6. Once upload is complete, a '☺' will appear on the laptop screen
 - 6.1. At this point, you are finished with the handheld
7. In EcoWatch, select the 'File' drop-down menu
 - 7.1. Select 'Open'
 - 7.2. Select the appropriate file name
 - 7.3. Select OK
8. Select 'File' drop-down menu
 - 8.1. Select 'Export'
 - 8.2. Choose 'CDF/WMF'
 - 8.3. Make sure that 'comma delimited' is selected in the export format screen
 - 8.4. Select 'Export'
 - 8.5. Now you are finished with EcoWatch
9. Open Microsoft Excel program
 - 9.1. Select 'File'
 - 9.2. Select 'Open'
10. Open the 'Ecowin' folder
 - 10.1. Open the 'Data' folder

- 10.2. Change the 'Type of File' menu to read 'All Files'
11. Select appropriate data file
 - 11.1. Text import wizard window will open
 - 11.2. File type should be 'Delimited'
 - 11.3. Select 'Next'
12. In the delimiters box, the 'comma' option should be selected
 - 12.1. Select 'Next'
 - 12.2. Select 'Finish'
 - 12.3. Data can now be saved in Excel

Appendix G- Algal testing

The following are instructions on how to accomplish algal toxin sampling. Only do this when advised by the field crew leader or manager.

1. Prepare pump for water filtering
 - 1.1. For an electric pump
 - 1.1.1. Plug in pump cord into boat power socket
 - 1.1.2. Attach one end of clear hose to pump, and the other to plastic Erlenmeyer flask
 - 1.1.3. Flip power switch to turn pump on
 - 1.2. For a manual hand pump
 - 1.2.1. Attach one end of the clear hose to pump, and the other end to plastic Erlenmeyer flask
 - 1.2.2. To pump, pull the hand pump to filter sample
 - 1.2.2.1. Use the pressure release valve typically located on the bottom of hand pump to release pressure
2. Sampling
 - 2.1. Gather 1L of surface water from sample location
 - 2.2. Separate cup from magnetized filtering stage
 - 2.3. Take out one algal toxin filter and place it on top of the filtering stage
 - 2.4. Replace cup on top, holding the filter in place and making sure the filter covers the whole area
 - 2.5. Place cup and stage inside the top of plastic Erlenmeyer flask, creating a tight seal
 - 2.6. Pour water in cup and turn on electric pump, or begin to squeeze hand pump to filter the water
 - 2.7. Continue pouring water in the cup until all 1L has been filtered
 - 2.8. Remove cup from stage, and take wet filter off, dry using a paper towel.
 - 2.9. Fold the dry filter in half and place it inside a small piece of tin foil
 - 2.10. Insert foil inside of a labeled coin envelope
 - 2.11. Place coin envelope inside a Ziploc bag. Place in coldest part of the cooler
 - 2.12. Dump filtered water and turn off pump