Standard Operating Procedure 14: Equipment Cleaning Procedures and Practices for Biosecurity, version 1.0

Background:
This document gives instructions for disinfecting gear that has come in contact with stream water and also provides some common sense measures to maintain clean gear in the field. This SOP was developed in direct response to increasing concern regarding the spread of *Didymosphenia geminate* (didymo) but pertains to other aquatic exotic species as well. Care must be taken to prohibit poor equipment hygiene, which can transfer exotics quickly as individuals/crews move between water bodies in the park. Additional care should be taken when gear is used outside park boundaries and then returned for use in the park. There are seven known didymo populations within 100 miles of Shenandoah National Park. Four of those are in free flowing streams (West Virginia, Maryland, and Virginia) and three are in tailwater systems (Virginia, Maryland). This SOP covers Fish Population Monitoring as well as Aquatic Macroinvertebrate monitoring and therefore, certain gear references may be irrelevant to any one particular protocol. Required equipment includes: 20 gallon muck buckets, buckets with lids, 30x26" trays, Dawn dish detergent, salt, water, hose, freezer, and drying rack.

In order to limit the introduction and spread of invasive or exotic aquatic organisms in Shenandoah, all equipment that is exposed to water is always cleaned prior to moving equipment between watersheds (i.e. Shenandoah, Rappahannock, James). Currently, with few exceptions, equipment is cleaned any time the crew moves between sampled streams (~5-6th field sub-watersheds).

Standard guidelines are to:

a) Use clean, disinfected gear in each stream (each site if practical)
b) Sample upstream sites prior to downstream sites when practical
c) Carry duplicate gear if needed
d) If practical don't use felt soled waders
e) Remove dirt and debris from equipment between sites
f) Dry buckets and tubs between sites
g) Clean/disinfect equipment at least daily if not more often (see below for appropriate cleaning solutions/steps)
h) Dry all equipment as often as possible
i) Packs will be hung, propped open and dried as long as possible between watersheds.
j) Buckets and tubs will be pulled apart each night and air dried. At the least, they will be disinfected in salt or soap solutions between major watersheds (James, Rappahannock, and Shenandoah River watersheds).
k) Sampling in streams can occur in up to four different watersheds in a single a day. All gear will be duplicated to the extent possible and one set will be soaked/cleaned while the crew is sampling a site with a "clean" set.
l) Ensure that when other crews borrow waders that they are responsible for cleaning

<u>Steps to Clean Gear</u>

1. **Choosing a staging area for cleaning gear**

Choose a fairly level area out of the way of traffic and other work groups that is close to a water source. This will ensure a safe place to work. In most cases, these areas are predetermined by the Biologist or Lead Technician.

2. **Washing containers**
   a) Two to three 20 gallon "muck buckets" are used for the salt solutions and one or two 30x26 inch trays are used for the detergent solutions.
   b) Measure out the amount of product you want for the volume of water needed and pour into the wash container. (A little more product is better than too little (see chart or directions below))
   c) Measure the amount of water needed and pour into the tub or use a tub that has been marked for this purpose so water can run straight in from the hose.

3. **5% Salt Wash (for use with non-metallic equipment)**

   Mix – Two cups of salt per 2.5 gallons of water or one pound salt per 5 gallons of water in one to two of the large colored tubs. Stir until salt is dissolved. A second or third tub should be onsite for rinsing. Waders, buckets, and other non-metallic gear will be soaked at least daily in this solution.

   a) Gloves should be washed in the salt solution first. They should be filled with solution individually, and then allowed to soak in the solution for **at least one minute** before draining and rinsing.
   b) The tapes, measuring trays, and other small items may be soaked with the gloves for **at least one minute.**
   c) Waders and wading boots should be scrubbed to get the dirt off and soaked in salt solution (**at least one** for non-felt items and **at least 30 minutes** for felt items). It is almost impossible to soak buckets and tubs, so they are rotated in the solution and allowed to sit for a couple of minutes before rinsing. The waders will have to be weighed down to stay submerged.
   d) Rinse items well with tap water and then put away to dry. Do not rinse equipment over soaking tubs.
   e) Rinse your hands well following exposure to the salt solution.

4. **5% Detergent Wash (for use with metal equipment)**

   Mix - 1 cup of Dawn or Palmolive dish detergent per gallon of water. Macroinvertebrate sampling equipment, nets, and probes will be disinfected with soap solution at least daily or air dried. *Note: Organic/green type detergents have been found ineffective at killing didymo so please use the specified products.*

   a) Metal gear should be soaked in dish detergent since salt is corrosive. Electrofishing probes, nets, measuring boards, depth sticks, Surber Sampler and non-felt waders should be soaked in detergent solutions for **at least one 1 minute.**
   b) It is almost impossible to soak buckets and tubs are rotated in the solution and allowed to sit for a couple of minutes before rinsing. (also see item "d" below)

c) The Portable Invertebrate Box Sampler (PIBS) and felt waders should be soaked for **at least 30 minutes**.
d) All soaked gear will be rinsed well with tap water and then put away to dry. Do not rinse equipment over soaking tubs. Buckets should be spread out upside down, not stacked.

5. **Air Drying** – To be effective, items must be air dried for 48 hours after they are dry to the touch. Buckets and tubs will be pulled apart each night and air dried. At the least, they will be disinfected in salt or soap solutions between major watersheds (James, Rappahannock, and Shenandoah River watersheds). Packs will be hung, propped open and dried as long as possible between watersheds.

6. **Freezing** – Can be used for any porous item, but space is limited and in most cases impractical.

7. **Use in the Field**
For cleaning equipment in the field 5 gallon buckets with lids can be used to store solutions. It takes about 4 gallons to adequately soak the Portable Invertebrate Box Sampler in the large black tray. Equipment should only be rinsed in stream water that the equipment is intended to be used in.

8. **Solution Degradation (i.e. how long can we use it?)**
Dish detergent will degrade with sunlight so the mixed solution should be kept no longer than a week. With care, the salt solution may be able to be used for as many as three weeks depending on how dirty the water gets. Additional water and detergent/salt will need to be added as the week progresses. Salt may harm grass over time even though the waders and other gear will be rinsed daily in the same location so chose an area with a harder surface for salt solutions while detergent may be used almost anywhere.

9. **Disinfection Options**– February 2010 (adapted from Biosecurity New Zealand/ National Institute of Water and Atmospheric Research Ltd.)

Treatment	**% Solution**	**Formula**	**Duration**	**Gear to treat**
Dish detergent (Dawn or Palmolive)	5%	1 cup per gallon of water	Greater than a Minute 30+ Minutes for felt	All gear
Salt	5%	2 cups per 2.5 gallon water OR one pound per 5 gallons water	Greater than a Minute 30+ Minutes for felt	Non-metallic items
Drying			Greater than 48 Hours	All gear
Freezing			Until frozen solid	Porous gear

Biosecurity Issues

- 5% Dish detergent is effective against Viral Hemorrhagic Septicemia (VHS), didymo, other aquatic plants, quagga/zebra mussels, New Zealand Mud Snails, and many other exotic aquatic species.
- 5% Salt is effective against all but VHS.
- Air drying of clothes is listed as 48 hours after material is dry to the touch.
- Freezing is listed as an alternative but space is limited and freezing time to "solid" is time consuming and most of our gear needs to be dry to use.
- 2% chlorine bleach is effective against all pests, but is extremely harsh on equipment for daily use and is toxic to aquatic systems. For these reasons, bleach is not considered to be a viable alternative.

SOP Revision History

All protocols, regardless of how sound they are originally, require modification as new and different information becomes available. SOP version numbers increase incrementally by tenths (e.g. version 1.1, version 1.2…etc.) for minor changes. Major revisions should be designated with the next whole number (e.g., version 2.0, 3.0, 4.0 …). Small changes or additions to existing methods will be reviewed in-house by Shenandoah or Mid-Atlantic Network staff. However, if a complete change in methods is sought, an outside review is required.

Revision History Log:

Prev. Version #	Revision Date	Author	Changes Made	Reason for Change	New Version #