S.O.P. FOR PFAS DREISSENID MUSSEL COLLECTION:

Extra Materials:

- Balance capable of measuring to 0.0 g
- Supplied pre-washed sample bottles and bags
- Cooler with ice or ice packs

*Implement the following S.O.P. at any sites where dreissenid mussels are clearly present in the wetland, clearly present adjacent to the wetland polygon or are collected via dip-net during regular CWMP sampling.

Note: Please take preventative measures by avoiding contact with surfactants (detergents, waterproofed fabrics, etc.) directly prior to mussel sampling events to minimize contamination.

Do not use gloves when collecting mussels.

Water Collection:

- If mussels are located in one of your CWMP sampling zones, the standard CWMP water collection and measures are sufficient (brief review below).
 - Composite water samples collected from the middle of the water column with an extension sampling tool to be analyzed ex situ for alkalinity, chlorophyll, and dissolved nutrients from filtered samples.
 - Collect surface in-situ water chemistry data using a multiparameter sonde at three replicates from each sample site.
- For sites where dreissenid mussels are collected outside of the wetland polygon, repeat the water sampling protocol both in the nearest adjacent coastal wetland as well as where collection occurs.

Mussel Collection:

- If possible, snorkel for mussel collection, but in the case where a sea wall or rip rap is housing dreissenid mussels, chest waders may be worn, and mussels can be handpicked or collected via dip net.
- Carefully remove mussels from any attached substrate while keeping the animal intact
- Collect at least 2 grams worth of whole dreissenid mussels from each replicate by hand for a total
 of 6 grams of tissue (absolute minimum, if organisms are smaller, collect 7-8 grams of organisms).
 It is recommended to bring a battery powered balance into the field to ensure minimum sample
 size. Shell accounts for approx. 65% of weight. 2 grams of tissue are needed for analysis.
- Place only whole mussels in pre-washed sample bottles, place bottles in pre-washed bags (do not
 place mussels directly in bags without being in bottles), label and store on wet ice or ice packs
 provided.
- Bag label should include the following: site name and number, date, replicate, crew leader name, and project name (i.e. PFAS project).

Following collection, store dreissenid mussel tissue samples in a -20 °C freezer until you are able to priority ship them to CMU's Wetland Ecology Lab.

S.O.P. FOR OXIDATIVE STRESS MUSSEL COLLECTION: Extra Materials:

- Access to a -80°C freezer and dry ice
- Styrofoam cooler with dry ice
- Regular Ziploc bags

Note: It is important that these mussels are collected after those being analyzed for PFAS because regular Ziploc bags are not an approved container for preventing PFAS contamination.

- In addition to the mussels collected for PFAS analysis, collect 4 dreissenid mussels by hand from 3 replicates (12 mussels total) from each sample site.
- Place extra mussels in standard Ziploc bags labeled with site name and number, date, replicate, crew leader name, and project name (i.e. PFAS/OS project).
- Place mussels in bags into a sealable polystyrene cooler full of dry ice.
- This will flash freeze driessenid mussels in situ. Samples must remain frozen until subsequent analysis.
- Following collection, store dreissenid mussels in a -80 °C freezer until shipment on dry ice to CMU's Wetland Ecology Lab.

IF YOU HAVE QUESTIONS/CONCERNS PLEASE CONTACT: CONNOR KOWALKE kowal1cj@cmich.edu (586) 530-4502