Arizona Game and Fish Department
Quagga Mussel Sampling Procedures
And Sample Collection Methods for PCR

Equipment Needed:

Plankton Sampler

Plankton net or sieve with 63 um mesh. The mesh size must be 63 um. The plankton net can have a 30 or a 50 cm diameter opening.

Ordering information for a 30 cm. diameter net from Wildco.

30-d28 with 63 um mesh	net
7-d30 --	ring and bridle
48--- c60	Adaptor 2.5 inch - -
47-d28	dolphin bucket
47-c99	guardrails

A plankton sampler can also be made from large diameter PVC pipe with a glued fitting to hold on the mesh. Nitex mesh can be purchased from Wildco.

Ethanol
Sample bottles – 500 and 1000 ml size
1 liter wash bottle
Ice chest
Decontamination container
Vinegar
Labels and sharpies
Substrate samplers
Rope

Pumps and hoses (canal samples)

Sample Methods:

The sample volume for PCR analysis is 1000 liters. Lake plankton tow samples should be a vertical tow. The tow speed is important so as to not exceed the filtering capacity of the mesh. Wash down the sides of the net using lake water and a wash bottle and collect the sample in the plankton net bucket. Transfer the sample in either a 500 or 1000 ml bottle, leaving enough volume for ethanol preservative.

Preserve the sample with ethanol. Ethanol volume is 25% of the total volume. The volume of the ethanol is not critical and can be estimated. When estimating ethanol volume, first mark a line on the bottle that shows the original sample volume and label (Level 1), add ethanol to the sample and label this volume (Level after ETOH). This is the method given by the Bureau of Reclamation lab so no need to worry that it is not that precise. An alternative method would be to have a large graduated cylinder to measure sample and preservative volume.

The sample bottle label should contain the following fields;

Sample date, time
Sample location, water body, site description, and GPS
Sample Depth
Volume of sample filtered through plankton net
Preservative
Sampler name and tel.

Transport samples in ice.

Collect water quality parameter information for pH, temperature, conductivity, and dissolved oxygen. Contact Marc Dahlberg to see if water samples for laboratory analysis should be collected

Equipment Decontamination

Sample equipment must be decontaminated after each site. Veligers will stick to the net mesh. Either a 5% acetic acid solution or vinegar will work to kill veligers and to denature DNA. The preferred method is to leave the net overnight in a vinegar bath, but a one hour contact time is sufficuieient to kill the veligers and to denature the DNA and prevent contamination to the following sample. Rinse the net thoroughly before processing the next sample. The same vinegar or acetic acid can be used repeatedly.

PCR Sample Shipping Information

Kevin Kelly/Denise Hosler (86-68220)
U.S. Bureau of Reclamation
Denver Federal Center
Corner of 6th Ave. & Kipling
Bldg 67, Room 152
Denver, CO 80225-0007

Contact information:
Kevin Kelly: kkelly@do.usbr.gov

Denise Hosler: Phone: (303) 445-2195; dhosler@do.usbr.gov
Fred Nibling: Phone: (303) 445-2202; fnibling@do.usbr.gov

Substrate Samplers

Substrate samplers can be paced at sample locations to document the establishment of adult quagga mussels in a waterbody. They can also be used to determine mussel population growth and seasonal abundance. Locate sample sites in an area of interest. Mussels are reported to prefer dark areas, crevice, undersides of boats.

Substrate samplers can be made from PVC, ceramic, or cement materials. PVC is the preferred plastic material. Acrylic and ABS plastics seem to not be a preferred material for adult mussel attachment. Substrate samplers should be left in place 2 months before being disturbed for monitoring. All substrate samplers should have an identification tag.

Substrate samplers can be attached to docks in a secure place. Sample locations that do not have some structure to attach to can be placed with a buoy with survey site signage and reflective material. Navigational Aids has same available materials and access to additional equipment.

PVC pipe samplers can be made from 2 inch grey pvc pipe cut into six inch lengths. Holes are drilled through the center of the pipe for rope attachment. 4 additional holes are also drilled into the sampler. A flat pvc samplers can also be used for substrate sampling This sampler is made from 1/8 inch pvc sheeting cut into 6 inch square pieces, which can be center drilled and attached to eyebolts for placement. Other commercially available substrate samplers are also available and can be used for monitoring of adult zebra mussels. If the sampler has a cement block or anchor attached this provides an additional sampling substrate and depth. Note the substrate type and depth with site information.

Quagga mussels seem to prefer habitat at lower depths. Samplers should be placed at lower depths within the hypolimnion, when there are sites of interest that have available depth to monitor this environment. Samplers should also be placed in the epilimnion to monitor this environment with sampler depth determined by lake transparency.

Sample Sites for PCR Analysis

Water	Location	Samples	Date
Roosevelt	Marina/Boat Ramp	1	
Canyon	Marina/Boat Ramp	1	
Saguaro	Marina/Boat Ramp	1	
Bartlett	Marina/Boat Ramp	1	
Pleasant	Marina and CAP discharge	2	
Salt River	Above Granite Reef	1	
Bubbling Ponds Hatchery	Outflow	1	
Willow Beach Hatchery	Inflow, Hatchery building outflow	2	
Alamo	Boat Ramp	1	
SRP South Canal	Below CAP outfall	1	
SRP Arizona Canal	Below CAP outfall	1	
CAP	Bouse Hills Pumping Plant	1	
CAP	Little Harquahala Pumping Plant	1	
CAP	Hassayampa Pumping Plant	1	
CAP	Waddel Pumping Plant	1	
CAP	Salt Gila Pumping Plant	1	
Colorado River	Imperial Dam	1	
Total		19	