# Early Detection Monitoring Manual for Quagga and Zebra Mussels



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# Early Detection Monitoring Manual for Quagga and Zebra Mussels

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This manual was developed to help direct early detection monitoring efforts for small lakes, reservoirs and streams in California that are believed to be free of invasive quagga and zebra mussels. The methods presented here are intended for citizen volunteer groups involved with or interested in monitoring aquatic organisms. Your work in helping identify the extent of the spread of these invasive species through early detection monitoring will be vital for minimizing the occurrence of additional infestations and for the application of effective treatment methods. While this manual addresses the situation in California, the majority of the information is broadly applicable to other states and countries. Be sure to consult your local fish and game authorities for current policies and monitoring needs.

Cover: Upper photograph: Pope Beach at South Lake Tahoe, free of invasive quagga and zebra mussels. Photograph by Tony Limas, El Dorado County. Lower photograph: Zebra mussels washed up on beach at Lake Erie. Courtesy of United States Environmental Protection Agency Great Lakes National Program office.

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# 1. Introduction

Aquatic invasive species (AIS), such as guagga and zebra mussels, are non-native aquatic organisms that have caused, or likely will cause, economic or ecological harm or impacts to human health. These species are often referred to as non-indigenous, nonnative, exotic, and alien species, though it is important to realize that not all exotic or non-native species are actually invasive or harmful. Often the word *nuisance* is used to express the harm they cause. Generally, those exotic species that are invasive have certain biological characteristics that allow them to reach high abundances, such as high reproductive rates or the ability to survive under stressful conditions. These characteristics allow them to thrive in new ecosystems, often causing economic and ecological impacts. AIS are recognized worldwide as pests that require specific management actions to prevent or minimize the impacts that result from their presence in an ecosystem.

AIS management actions typically include three components, with associated education and outreach:

- Prevention
- Early Detection
- Eradication/Control

Preventive measures are designed to minimize the likelihood of introduction and subsequent establishment of an AIS to an area. Preventing the introduction of an AIS is typically the most cost-effective and best management approach. If the pest doesn't become established, it doesn't have to be dealt with. Preventive strategies vary with the type of water body being protected and the type of organism and vectors being addressed. While prevention is the most desirable management option, it is often difficult to fully protect a water body from introductions of AIS. This is particularly true for very small organisms that are not easy to detect. Thus, early detection monitoring is implemented to complement preventive measures.

Early detection monitoring is used to evaluate the presence of an AIS in a water body. Such detection is critical for many reasons. First, if the population is detected early, before it gets too widely distributed, eradication strategies can be considered. *Early* doesn't necessarily mean when it first arrives, rather that the established population hasn't had time to explode and become widely distributed. The time necessary for an established population to become highly abundant varies tremendously from system to system, and in some cases it may not happen at all. The likelihood of eradication also is influenced by the type of organism and its associated life cycle. But, the sooner a population is detected, the more time there will be to take action and the higher the likelihood of successful eradication. Responding to an infestation at an early stage is also referred to as rapid response. Rapid response plans for AIS in general, and guagga/zebra mussels in particular, are being developed and updated in California (Appendix I).

Early detection monitoring is also vitally important for minimizing the spread of the AIS to other places. As has been done with the quagga mussel in Southern California, additional actions are taken to protect other water bodies once an infested water body has been identified. If we don't know which water bodies are sources of the AIS, the AIS will likely continue to spread. With early detection, we can identify the sources and implement actions to prevent additional spread of the AIS. However, it is essential that those involved in early detection take appropriate steps to decontaminate themselves and their gear so as not to be a vector for spreading AIS while participating in monitoring activities.

Once an AIS has been detected, eradication or control programs are typically necessary. Development of these post-detection management programs requires more indepth monitoring. Post-detection monitoring uses different, more standardized methods than early detection monitoring, and it provides information on the distribution and abundance of the various life history stages of the AIS. This type of monitoring is also used to measure the efficacy of implemented eradication and control measures. Carefully designed, scientifically based eradication/ control programs can eliminate or minimize both economic and ecological impacts of the AIS. Eradication and control strategies often involve one or more physical, mechanical, chemical or biological methods.

While eradication of AIS has proven to be difficult for most species, success is marked by timely implementation and a sound scientific design for the targeted system. Great strides have been made over the last several years in developing effective eradication programs for AIS, with more successes now occurring. Even if eradication isn't attainable, control of the population is still critical.

Control programs can be highly effective for decreasing the impacts of the AIS on the infested ecosystem and may help reduce the likelihood of spread into uninfested habitats. As with eradication programs, control programs need to be well designed and developed for the specific infested water body and the target organism, with an implementation and funding structure that allows for long-term management. Evaluating potential control strategies for a specific water body prior to detecting a pest population enables a quick response if eradication efforts do not work. In such cases, early detection is still essential for minimizing the chance for a large increase in abundance of the pest population.

The combination of prevention, detection, control with a variety of appropriate measures and monitoring effectiveness can be referred to as integrated pest management (IPM). Because implementation of these strategies can impact native species and habitats, it is important to carefully evaluate the options doing certain things can be worse than doing nothing!

### **This Manual**

Quagga (Dreissena bugensis) and zebra (Dreissena polymorpha) mussels — also known as Eurasian mussels because of their place of origin, as well as Dreissenid mussels because of their common genus — have caused numerous economic and ecological impacts where they have been introduced. Since the late 1990s, California has conducted many prevention programs to keep these pests from entering the state's water bodies. Some of these programs (e.g., boat inspection trainings, outreach/education programs) are still available to groups, organizations and/ or the public, but they are not covered in this manual. For more information about and resources for inspection and prevention see Appendix I.

Unfortunately, prior to the implementation of some prevention methods, guagga and zebra mussels became established in several water bodies. Nonetheless, it remains important to reduce the possibility of spread to additional water bodies by encouraging and participating in the cleaning of all watercraft (e.g., motor, sail, kayak) moving among water bodies (Appendix I). There is also an essential need to determine which additional water bodies may be infested and which are still free of the pests. Volunteer early detection monitoring programs that are coordinated with water and resource agency programs are a cost- and time-effective way to evaluate the presence/ absence of AIS in water bodies. Such programs provide more eyes looking for the mussels at a fraction of the cost of agencybased programs.

This manual was developed to help direct early detection monitoring efforts for small lakes, reservoirs and streams in California that are believed to be free of quagga and zebra mussels. The methods presented here are intended for citizen volunteer groups involved with or interested in monitoring aquatic organisms. Importantly, early detection monitoring is designed to provide a measure of presence/absence, but typically does not include quantitative measures of abundance. More in-depth monitoring requires different techniques and biological monitoring expertise.

This manual includes four primary sections:

- Background
- Sampling Considerations
- Survey Methods
- Decontamination

The first section, Background, reviews the history, biology, identification, spread, impacts and management of guagga and zebra mussels in the United States. The second section, Sampling Considerations, describes information to consider when developing a monitoring program, including what life stage to sample, when and how often to sample, and where to sample. The last two sections, Survey Methods and Decontamination, provide details on the necessary monitoring components and procedures for monitoring guagga and zebra mussels and other AIS. We have also included four appendices. Two of these provide additional information sources and contacts. The other two — Quick Reference Sheet: Generalized Monitoring Procedures and Data Sheets — are for use in the field. Collectively, this information is intended to help individuals and groups understand the need for early detection monitoring and assist with the development of a monitoring program that fits their needs and abilities. If you are interested in monitoring a water body for guagga and zebra mussels, we encourage you to contact your local Fish and Game authorities to coordinate your efforts with those of others.

There is no doubt that the quagga and zebra mussels are tough non-native species! But, with proactive measures — including early detection monitoring and planning — we will be better prepared to minimize the impacts of these pests in California and elsewhere. We hope this manual facilitates more efforts toward managing these and other AIS.

# 2. Background

The quagga mussel, *Dreissena bugensis*, is native to freshwater areas of Ukraine. The zebra mussel, *Dreissena polymorpha*, is native to Ukraine and the low salinity areas of the Caspian, Black and Asov seas in southeastern Europe and western Asia.

Quagga and zebra mussels were originally introduced into the United States in the 1980s when transatlantic ships released infested ballast water into the Great Lakes. These invaders spread throughout the Great Lakes system, then throughout the central and northeastern U.S., through the Mississippi drainage and later into the Southwest. In the 1990s, zebra mussels were occasionally found on trailered boats entering California. By January 2007, quagga mussels were found in Lake Mead, a reservoir on the Colorado River along the Nevada/Arizona border. Researchers believe the population may have been established as many as four years earlier. Since 2007, these mussels have spread throughout the Colorado River, the Colorado River Aqueduct, and reservoirs fed by that system, including several water bodies in Southern California. The first established population of zebra mussels in California was detected in January 2008 at San Justo Reservoir in San Benito County. Both guagga and zebra mussels may be present in other water bodies in California, but remain undetected. Maps showing the distribution of quagga and zebra mussels in California are available at the Department of Fish and Game's Quagga and Zebra Mussels webpage at www.dfg.ca.gov/invasives/guaggamussel. For daily updated maps of the current distribution of guagga and zebra mussels in the United States, visit the U.S. Geological Survey's Nonindigenous Aquatic Species *Program* website at *http://nas.er.usgs.gov/* taxgroup/mollusks/zebramussel.

### 2.1. Biology Basic Ecology

Mussels in the family Dreissenidae, including quagga and zebra mussels, are relatively small (< 2"; < 50 mm) freshwater bivalves, having two hinged shells (or valves) that surround the body. They breathe and feed by inhaling water and passing it over their gills, removing oxygen and microscopic plants and animals (plankton). Quagga and zebra mussels have incredibly high filtration rates; an adult mussel is capable of filtering almost one liter of water per day.

### **Reproduction and Development**

Quagga and zebra mussels have separate sexes and reproduce by broadcast spawning, where eggs and sperm are released into the water column (Fig. 1). A mature female is capable of producing a million or more eggs per year. In their native habitat, spawning peaks in spring and fall, but in the warmer waters of western North America, they may spawn year-round. After fertilization, the embryos develop into free-swimming planktonic larvae called veligers (Fig. 2). Veligers are planktonic, meaning they live unattached, floating in the water column (Fig. 3). They have a small, rounded shell, and a flap of soft tissue, called a *velum*. This structure is fringed with sticky little hair-like structures (*cilia*), which grab food particles and move them to the mouth. Veligers of both quagga and zebra mussels are microscopic, ranging in size from 39–500 microns (0.0039–0.05 mm). These veligers remain in the water column until they are more developed and find a suitable place to settle. This can take from 5-240 days, though most settle out within six weeks. At this point, they metamorphose into a juvenile that resembles a small adult and settle out of the water column. The mussels then use



**Figure 1.** *Dreissena* spp. life cycle. Illustrated by Chris Webb, U.C. Cooperative Extension. Adapted from *http://www.fws.gov/Midwest/mussel/images/zebra\_mussel\_%20life\_cycle.html*.

their foot to crawl along potential settlement sites and assess whether the substrate is suitable for attachment. If the substrate is desirable, they will secrete byssal threads (small thin fibers) from their foot and attach to the substrate. If the site is not suitable, they will continue to crawl on the surface of the substrate or they will pull up their foot, crawl or drift to another substrate, and start the process over until they find a preferred substrate.

Young juvenile quagga and zebra mussels may first attach to filamentous substrates, like aquatic weeds (Fig. 4). Mussels that settle on these substrates are typically quite small (1/32–1/8"; 1–3 mm). As the mussels grow (and the fibrous plants naturally decay) they may release their byssal threads and move to a hard substrate where they often stay for the remainder of their lives. Benthic mussels are those whose attachment sites are found at the bottom of a water body, typically in association with hard substrates like rocky habitat or shelled organisms. However, guagga and zebra mussels are also often found attached to hard substrates in dark places in the water column, even near the surface, such as the underside of docks. If environmental conditions change such that the area or substrate is no longer suitable for the mussel, it can detach and relocate. Quagga and zebra mussels grow and mature

within one year, and potentially much sooner in the warmer waters of the Southwestern United States.

### Habitat Requirements

There have been few studies of the habitat requirements and tolerances of guagga and zebra mussels in the Western United States. Thus, it is unknown whether these factors differ from the known parameters for these mussels in other regions. Based on studies from the American Great Lakes region, guagga and zebra mussels have similar environmental requirements (Table 1). Both require conditions where salinity is low, calcium (needed to form shells) is relatively high, pH is somewhat alkaline and water flow is slow. They avoid brightly lit areas, preferring darker, shaded areas. Both species prefer to attach to hard substrates. However, they are also

found on softer substrates such as sand or aquatic plants; this is more common for the quagga mussel particularly when it becomes abundant or reaches high densities in an area. Because they can close their shells fairly tightly, mussels of both species can survive out of water from five days to one month, depending on temperature and humidity. If kept out of water they will



**Figure 2.** Generalized mollusk veliger. Illustrated by Chris Webb, U.C. Cooperative Extension. Adapted from USACE drawing, Hopkins 1990.



**Figure 3.** *Dreissena* spp. life history. Note, while approximate scale bars are included for the magnified views of plankton and benthic mussels, other elements are not to scale. Illustrated by Chris Webb, U.C. Cooperative Extension. Adapted from *http://www.fws.gov/Midwest/mussel/images/zebra\_mussel\_%20 life\_cycle.html.* 

eventually starve or dry out, which happens more quickly in warmer and drier conditions.

Generally, quagga mussels have broader habitat and environmental tolerances than zebra mussels (Table 1) — for example, they can handle lower water temperatures. In addition, zebra mussels are generally found

in shallow water, whereas quaggas can be found at much greater depths as long as there is sufficient oxygen. Further, both guagga and zebra mussels can survive at fairly low oxygen levels, but they apparently require at least 25% oxygen saturation to grow and reproduce. Some researchers speculate that guagga mussels can withstand lower oxygen levels than zebra mussels, but currently there is no definitive proof.

### Predators

In their native range there are several natural predators of these mussels, including aquatic birds and fishes such as catfish and sunfish. In North America, diving ducks, red-eared sunfish, carp, drum, and a few catfish species are known to feed on quagga and zebra mussels, although they have not yet been observed to control rapidly multiplying populations.

# 2.2. Identifying Quagga and Zebra Mussels

When first undertaking a monitoring program, identifying quagga and zebra mussels can be challenging because they can be confused with other animals, plants and even nodules of rust. Mussels are most readily distinguished from other organisms by the way they are attached to a substrate. Because mussels use byssal threads to attach, they will rotate and stay attached when pushed on. This is true even for very small mussels. Most other animals or objects will fall off (e.g., snail, clam, pebble), be pushed

to another location on the substrate (e.g., limpet), or will remain fixed (e.g., encrusting organisms) without any rotation.

In California, quagga and zebra mussels (Figs. 5 and 6) are most often confused with the Asian clam, *Corbicula fluminea*, also an



**Figure 4.** Juvenile mussels (indicated by arrows) attached with byssal threads to natural vegetation. Photograph by Charles Ramcharan, Wisconsin Sea Grant.



Figure 5. Comparison of shell morphology of zebra and guagga mussels. Photograph by Myriah Richerson, USGS.

invasive non-indigenous species in many areas (Fig. 7). However, this species can be easily distinguished by its lack of attachment and different shell features (Appendix IV). Like mussels, the Asian clam also has two shells (thus, is a 'bivalve'), but it does not attach to hard substrates, burrowing into sand or mud instead. In addition, they have symmetrical shells that are typically all one color — light to dark brown, although there may be a white patch near the hinge (bottom) of the shell, and their shells have noticeable ridges. Snails or limpets are other mollusks that, like mussels, may appear to be attached to the substrate, but these organisms do not have two hinged shells and they do not attach using byssal threads — they have a fleshy foot like a garden snail. One snail that many people have heard about, and may encounter

when monitoring, is the New Zealand mudsnail — another AIS. These animals are easy to distinguish from mussels: they have a single, spiraling shell, are not firmly attached to the substrate, and adult New Zealand mudsnails are tiny compared to adult guagga and zebra mussels (Figs. 8 and 9).

It is also possible to confuse quagga and zebra mussels, especially the small

Quagga Mussel Zebra Mussel Dreissena polymorpha Dreissena bugensis Obvious ridae Lacking ridge Bilaterally symmetrical. Joined together in a midventral line. Byssal Groove Byssal Groove Asymmetrical, no straight midventral line.



juveniles, with buds and spores of plants and algae, fish or amphibian eggs, or even inanimate objects like rust nodules. To avoid this, confirm that the object has two hard shells (not gelatinous) and has threads attaching it to the substrate. We recommend consulting with local water body staff biologists and others knowledgeable about local

freshwater organisms prior to beginning a monitoring program to learn more about the species you may encounter. They may even have samples of local invertebrates you can examine.

Importantly, while distinguishing mussels from other organisms is critical for the purposes of early detection monitoring, it is not important to distinguish between quagga and zebra mussels. Both species are problematic, and they can be difficult to tell apart because they are only slightly different in appearance.

If you are interested in differentiating between guagga and zebra mussels for your own purposes, one key distinguishing characteristic is the shape of the hinged-side

Parameter	<b>Zebra Mussel</b> Dreissena polymorpha	<b>Quagga Mussel</b> Dreissena bugensis
Salinity	< 5 parts per thousand	< 5 parts per thousand
Calcium – optimal	> 25 mg/L*	> 25 mg/L*
рН	7.4–9.5	7.4–9.5
Water velocity	< 2 m/sec (6.6 ft/sec)	< 2 m/sec (6.6 ft/sec)
Substrates	Prefer hard substrates	Prefer hard substrates, but may be found on softer substrates
Water depth	Surface to 55 m (~ 130 ft) prefer depths 4–12 m (~ 13–40 ft)	Surface to > 120 m (~ 400 ft), prefer depths < 30 m (~ 100 ft)
Temperature – range for survival	>0-30° C* / 32-86° F*	5–30° C* / 41–86° F*
Temperature – optimal for reproduction and growth	64° F / 18° C	60° F / 16° C
Temperature – minimum for reproduction	12°C/54°F	9° C* / 48° F*
Oxygen – optimal saturation	25 % saturation	> 25 % saturation*
Oxygen – minimum required for survival	2 mg/L	>2 mg/L*

Table 1. Physical habitat requirements for quagga and zebra mussels.

Derived from data from their native range and the American Great Lakes (Britton 2007; Cohen 2007). Some parameters remain unstudied and may be different in the Southwestern United States, where environmental conditions differ. \*Exact number is unknown.

of the shell (Figs. 5, 6). Zebra mussels have a broad, flat edge along the hinge, and as a result they will usually stand upright when placed on this edge. In contrast, the hingedside of the guagga mussel shell is guite thin, more rounded and lacking a distinct flat edge. Consequently, the guagga mussel will roll over to one side, as it is unable to stay upright on its hinge. In addition, the hinge itself is curved on the guagga mussel, but straight on the zebra mussel (Fig. 6). Though color is highly variable, zebra mussels often have more pronounced, darker, thicker and jagged stripes, while quagga mussels have thin stripes that are typically more pale in color. But again, keep in mind that distinguishing between these species is not important; determining whether it is a mussel is crucial.



**Figure 7.** The Asian clam, *Corbicula fluminea*, an invasive non-indigenous species in California and other regions. Photograph by Noel Burkhead, The Nature Conservancy.



**Figure 8.** Adult New Zealand mud snail (left) and juvenile quagga mussel (right). Adult mussels are much larger than the juvenile mussel here. Scale shown in inches. Photograph by Matt Newnham.

originally introduced adult quagga mussels to the Colorado River system, the subsequent spread into California resulted from the transfer of mussels through the water supply infrastructure.

In addition to these primary means of spread, some have suggested that quagga and zebra mussels might also be spread via wildlife, attached to mobile crustaceans (Fig. 10), waterfowl or the fur of mammals. This mode of spread appears to be insignificant when compared to trailered watercraft and interbasin water transfers.

### 2.4. Impacts

Both quagga and zebra mussels cause substantial ecological and economic damage.



Figure 9. Adult New Zealand mud snail. Photograph by Jane and Michael Liu, Oregon Sea Grant.

### 2.3. Vectors of Spread

Because quagga and zebra mussels can survive out of water for weeks, adults and juveniles can easily be spread between water bodies by boats, trailers and other equipment where they are attached. The mussels attach in nooks and crannies under the waterline on boats and engines, as well as to damp objects such as coiled ropes in storage areas of boats. Further, the planktonic veligers can be carried to new locations in any standing water, such as the water left in the bilges, engines and drywells of boats.

When water conveyance systems (e.g., pipelines, aqueducts) connect two water bodies, they form another potential avenue, or vector, to spread adults or veligers. While it appears that a recreational boat may have



**Figure 10.** Crayfish covered with juvenile zebra mussels. Photograph by Ontario Ministry of Natural Resources, courtesy of GLSGN Exotic Species Library.

### Ecological

Given their ability to filter large volumes of water, in combination with reaching extremely high densities, the quagga and zebra mussels significantly reduce the amount of suspended particles (including plankton — food) in the water column. As a result, the ecosystem is altered because there is increased water clarity and less food available. In addition, both quagga and zebra mussels are able to accumulate organic toxins in their tissues at more than 3,000 times the concentration in the environment. When animals feed on mussels that contain the toxins, the toxins



**Figure 11.** Boat propeller encrusted with quagga mussels. Photograph courtesy of the National Park Service.

are passed up the food chain, increasing wildlife exposure to pollutants. Quagga and zebra mussels will also colonize beds of native clams and other invertebrates. Subsequently, the feeding abilities of the colonized native organisms are hindered, often to a point of greatly stressing and killing individuals and possibly impacting populations. A potential secondary effect caused by increased water clarity, and therefore increased light penetration, is the growth of aquatic weed infestations.

### Economic

Both guagga and zebra mussels cause great economic damage to infrastructure, including water supply and water-based cooling systems, as well as docks, breakwaters, buoys and other marina infrastructure and vessels themselves (Fig. 11). The mussels' ability to colonize on hard surfaces can clog pipes, screens, valves, etc., costing hundreds of millions of dollars in repairs and maintenance every year. In addition, when the mussels decay, they corrode steel and cast iron pipelines that must then be replaced. The added cost to maintain water supply systems that have become infested with guagaa and zebra mussels may be passed on to consumers. Indeed, this already has happened to some customers in Southern California.

# 2.5. Current Management Actions in California

There are many ongoing management activities in California being carried out by

federal, state and local agencies, water suppliers and other partners interested in limiting the ecological and economic harm caused by guagga and zebra mussels and other AIS. Activities include a combination of prevention, early detection, containment, monitoring and, where possible, eradication and control (see Appendix I: General *Management*). Several agencies have participated in extensive outreach and education efforts. One agency, the California Department of Food and Agriculture, operates border protection stations that conduct vessel inspections at points of entry to the state. Other agencies conduct monitoring using surface surveys, deployed artificial substrates, plankton samples and divers. The monitoring methods described in this manual are some of the same methods being used by various agencies.

There are also ongoing projects exploring new management techniques, such as using dogs to detect the scent of invasive mussels, applying special paints and coatings to infrastructure, and the use of biological control agents. Your work in helping identify the extent of the spread of these AIS through early detection monitoring will be vital to the application of effective treatment methods.

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# 3. Sampling Considerations

When developing an early detection monitoring program for quagga and zebra mussels or other AIS, there are a variety of factors to consider. These include monitoring goals, life cycle stage, surveying period, site selection and permits and permission.

### **3.1. Monitoring Goals**

The goal of a monitoring program is one of the first considerations in determining how, when and where to monitor. In this manual, we focus on detecting an infestation at its earliest stages to determine whether or not the AIS is present. Other reasons to monitor might be to evaluate the spread of an infestation throughout a water body, or the success of an eradication or control program. In these cases, the AIS has already been detected and a more comprehensive monitoring program is required to manage the invasive population. Post-detection monitoring is not discussed in this manual.

### 3.2. Life Cycle Stage

Many aquatic organisms have complex life cycles involving several different stages (e.g., larval, juvenile, adult) that often occur in different habitats. It is important to consider the life cycle stage that will be targeted to develop an effective monitoring program. Not only are different methods and equipment required for monitoring different stages, but the sampling period and frequency likely will differ (see section *4. Survey Methods*).

For example, to determine whether quagga or zebra mussels are entering a water body from a particular water source (piped-in or truckedin water), monitoring for the larval stages in the water column should be conducted. However, if boaters are suspected as the primary source for introduction, then juvenile and adult mussels should be surveyed by monitoring attachment substrates. While boats can be a source of larval mussels, larvae typically settle out of the water column within a few weeks (depending on water temperature), making them difficult to detect unless they are continually being introduced. Decide which stage(s) will be monitored and then use appropriate methods (see section *4. Survey Methods*).

### 3.3. Surveying Period and Frequency

To determine when and how frequently to monitor for AIS it is necessary to understand the biology of the targeted life stage, as well as the influence of environmental parameters on the organism being monitored. Water temperature typically influences when monitoring occurs, because it affects the timing of reproduction (spawning) as well as the growth and development of organisms. If looking for early life stages of an organism, an early detection program should coincide with the spawning season. For juveniles and adults, it usually is easiest to detect these stages after the peak spawning period and when the organisms are growing rapidly.

The timing and frequency of surveys often differs according to climate. For example, because water temperatures in low-altitude water bodies of Southern California enable quagga mussel reproduction throughout the year, monitoring should occur year round. In areas further north or at higher altitudes where temperatures vary significantly in different seasons, mussel reproduction and growth will likely cease or greatly slow. Thus, monitoring may be intensified in warm summer months and suspended during cold winter months.

In many cases, the frequency of monitoring is determined by the amount of time that a

volunteer has to participate in the activities. However, the biology of the target organism should always be taken into consideration. It is most productive to have a consistent schedule, such as weekly, biweekly, monthly, quarterly, or annually. Certainly, the frequency may be increased during high-risk months (i.e., when there is a high likelihood of introduction due to increased vector activities, or during the optimal reproductive and growing season). If only a limited amount of time can be expended on an early detection monitoring program, monitoring efforts should occur during the high-risk periods. For guagga and zebra mussels, this would be during months when boating or water supply pumping activities are increased and introduction of mussels is most likely, or during months when the water is warm and optimal for mussel spawning and growth (Table 1). Coordination of monitoring efforts by all groups working at a water body is recommended so that an optimum sampling strategy can be implemented throughout the year.

### 3.4. Site Selection

There are several key components to take into account when selecting early detection monitoring sites to maximize the success of the program:

- Environmental parameters
- Proximity to high-risk areas
- Site accessibility
- Safety

Local water body staff should be engaged in early discussions of potential monitoring activities. They can provide useful information about the water body that facilitates development of a safer and more effective early detection program. Also, they can coordinate your activities with other ongoing monitoring efforts.

### **Environmental Parameters**

There are two primary environmental factors to consider when choosing early detection monitoring sites for quagga and zebra mussels: 1) light level and 2) water current (see section *2. Background*).

### Light level

Quagga and zebra mussels prefer low light levels, and thus are found in shaded and deep water. Early detection monitoring sites should take advantage of this trait and target locations with low light levels.

### Water current

Quagga and zebra mussels also prefer areas of low flow (1–2 m/sec). Sites with slow moving water or eddies (where water slowly swirls around in the same location) are good early detection monitoring sites. For streams, the best locations may be along the edges of runs and pools where the water currents are typically slower.

### **Proximity to High-Risk Areas**

High-risk areas are locations where the AIS are most likely to be introduced to a water body, and are where monitoring efforts should be focused. Based upon the main vectors of introduction of quagga and zebra mussels, there are three primary types of high-risk areas:

- Water inflow areas and structures (pipeline discharge, water entry points)
- High-use boating areas
- Substrates with substantial surface area (docks, boat ramps, pipelines, floating bathrooms)

Locations at and down-current from either water supply entry points or high-use boating sites are high-risk areas since the mussels are likely to be introduced there and may drift down-current. This includes areas with boat ramps and docks, particularly those used by non-resident vessels as they are a likely source of introduction. Areas with substrates having a large amount of surface area (e.g., docks, pipelines) are also key monitoring locations, because if mussels are present they likely will end up attaching to these large surfaces.

### Accessibility and Safety

Easily accessible, safe areas should be a top priority when identifying potential early detection monitoring sites. Special care should be taken when working in high traffic areas to minimize interference with boating, swimming, fishing and other water activities. General safety in terms of water and road conditions, as well as remoteness should be evaluated for each potential site. Some areas can become slippery or experience flash flood conditions when it rains. Remember to carry a cellular telephone, especially when working at remote sites, and use the buddy system. If another person cannot accompany you, make sure someone knows you are out monitoring and check in with them upon your return. While some sites may be ideal for mussels. they may be unsafe for fieldwork and thus should be excluded from consideration. Always consider your safety and that of other volunteers first. Also, be sure to consult with those people managing the water body about potentially hazardous locations.

### 3.5. Permits and Permission

Before you begin surveying, it is imperative to obtain the necessary permits, legal access and permission to monitor a water body.

### Permits

No permits are required to examine existing and deployed substrates. However, collection of any organisms or substrate suspected to harbor mussels or any other organism is prohibited by Fish and Game Code Section 1002 and 2301, and Title 14 Sections 650 and 670.7 (*http://www.dfg.ca.gov/wildlife/ species/regcode.html*) without the appropriate permit. For information on current permit requirements, please contact the California DFG regional quagga/zebra mussel staff (Appendix II) or your local fish and game authorities.

### Permission

Prior to initiating a monitoring effort at a location, permission will be required from the organization responsible for that water body. Additional permits or liability waiver releases may be required by the water districts and other agencies associated with the monitoring sites. Public lakes and reservoirs are often administered by a government agency, while private water bodies may be administered by a homeowners' association, board of directors or individual. Often the park ranger, lake recreation manager or general manager are the best people to speak to first. These people can bring requests to advisory boards or, if necessary, the California Department of Water Resources or the U.S. Bureau of Reclamation.

# 4. Survey Methods

### **Recommended General Field Gear**

Early detection monitoring team members need to be equipped to handle dirty and wet field conditions, exposure to sun, biting insects, and depending on the site, rough vegetation (including poison oak) and debris. We recommend having the following gear when conducting your surveys:

- Closed-toed shoes with good tread
- Long-sleeved shirt and long pants
- Hat or visor
- Sunglasses
- Sunscreen
- First aid kit
- Disinfectant hand wash
- Buddy and/or contact person
- Digital camera
- Cell phone
- Water and snack

### Approaches

In general, three survey approaches are used to monitor quagga and zebra mussels:

- Existing surfaces
- Artificial substrates
- Plankton

These approaches target different life stages, have varying sampling regimes, and require differing amounts of labor and sample processing costs (Table 2). Overall, surveys of existing substrates are least expensive, and plankton surveys are the most expensive. Since all of these methods have limitations, combining techniques is the most effective means for detecting the presence/absence of mussels in a water body. Section 4.2. *Artificial Substrate Surveys*, explains when and how to augment surveys of existing surfaces with artificial substrate surveys.

Survey Type	Life Cycle Stage	Optimal Sampling Frequency	Optimal Sampling Period*	Equipment Costs	Labor Required	Sample Processing Costs**
Existing Surfaces	Juveniles Adults	Twice Monthly	Year Round (warm climates) May-October (seasonal climates) June-September (high-risk months) <sup>1</sup>	Minimal	\$	n/a
Artificial Substrates	Juveniles	Monthly	Same as Existing Surfaces	\$\$ (substrates/ lines)	\$\$	n/a
Plankton	Veliger Larvae		Same as Existing Surfaces	\$\$\$-\$\$\$\$ (net/ microscope)	\$ <b>-</b> \$\$\$\$ <sup>2</sup>	\$\$\$

Table 2. Sampling methodology selection considerations.

\*Sampling period will vary depending on environmental conditions of water body.

\*\*Does not include the labor associated with processing the samples.

<sup>1</sup>Months with a high likelihood for introduction and detection (see section *3.3. Surveying Period and Frequency*).

<sup>2</sup>Major amount of labor required if samples are examined via microscope or molecular methods.

While the following sections are quite detailed, Appendix III provides a summary of the recommended survey methods. This appendix can be copied and referred to while in the field.

### 4.1. Existing Surface Surveys

There are a variety of methods for surveying existing surfaces within a water body: visual, tactile and underwater (diving). Existing surfaces include any substrates already in the water such as boats, docks, buoys, anchors, rocks, sand, or organisms (e.g., shellfish, aquatic plants). Many of these substrates are composed of hard materials such as cement and wood that are ideal for mussel settlement. Monitoring existing surfaces is a good activity for volunteer groups since it is simple and cost-effective. However, some existing substrate survey techniques require specific expertise (e.g., SCUBA certification) or field equipment (e.g., ponar grabs, metal box scrapers, coring devices). Here we focus on techniques that can be implemented without special equipment or intensive training, although we've also included brief information on in-water dive and snorkel surveys.

Before starting, remember to obtain the necessary permits and permission (see section 3.5. Permits and Permission). Special clearance and liability waivers may be required for examining docks and pulling up buoys or lines. Do not touch or collect samples from privately owned boats, buoys, lines, traps, or other property without permission. In addition, monitoring activities may have to be scheduled at times when there is less traffic and activity at the water body. Water body staff should be consulted when determining scheduling. Always be respectful of private property when conducting early detection monitoring activities.

### **Targeted Life Stage(s)**

Surveys of existing surfaces are useful for assessing both settled juvenile and adult mussel populations. A combination of visual and tactile (by touch) surveys can be used to detect mussels larger than 1/8" (3 mm) in shell length — a size at which the mussels are visible to the unaided eye. Tactile surveys are required for recently settled small juveniles that are less than 1/8'' (3 mm) in shell length.

### **Surveying Period and Frequency**

It is ideal to survey existing surfaces once every two weeks. It is better to have an intense monitoring effort bi-weekly, or even monthly, than a less intensive effort more frequently. Conversely, it is better to have more frequent monitoring efforts spaced two weeks to one month apart rather than more intensive efforts just a few times a year. Remember, if only a limited amount of effort can be expended for such surveys, target the months when the water temperature is optimal for mussel spawning and growing (Table 1). Also coordinate your efforts with other groups to ensure an optimum sampling strategy for the water body.

### **Site Selection**

Surveys of existing surfaces are best accomplished where the surfaces are: 1) easily viewed and/or reached, 2) located in areas having low light and slow water currents and 3) permanent structures that are at high risk of infestation (e.g., boat ramps, buoys and moorings). Based on all of these parameters, we recommend surveying the following surfaces:

- Underside of docks
- Shaded side of permanent structures (e.g., dock pilings)
- Underside of navigational buoys, moorings and other similar substrates
- Inside crevices of anchors and other substrates
- Natural materials (rocks, wood, aquatic plants and animals)

The first four surfaces mentioned above are typically surveyed, but don't forget to examine natural surfaces as well, including plants and even other animals. The mussels can exist under a layer of organic debris or fine sediment, attached to or shaded by aquatic plants or even connected to the surface of other shellfish.

### Visual and Tactile Surveys

Quagga and zebra mussel populations can be surveyed along the length of almost any surface or shoreline. Some potential mussel settlement substrates can be viewed from the



**Figure 12.** Visual inspection of dock undersurface. Photograph courtesy of California Department of Water Resources.

surface (docks) (Fig. 12) or, as in the case of buoys or anchor line, may be pulled up and inspected on a boat, dock or from shore. Look very closely for these animals and inspect all types of surfaces on the shaded portion of the substrate. While minimum search distances are recommended in Table 3, it is best to search as much surface area as possible.

**Table 3.** Minimum surveying distances for visual surveys of various surfaces (Source: DWR Zebra/ Quagga Mussel Surface Survey Protocol).

Surface	Minimum Sample Size (in Feet)
Boat Ramp Bottom in a Marina	100
Boat Ramp Bottom (solitary)	200
Shoreline	100
Mooring/Dock Lines	200
Anchor/Dock Cable or Chain (portion under water)	100
Concrete Structures	100
Logs and Woody Debris	100

Tactile (by touch) surveys can be used to detect small juvenile mussels on surfaces that can be reached but not easily viewed. To inspect tactilely, simply run a bare hand along the length of the surface. Be careful and use a light touch since mussels, as well as other organisms living on the substrate, can have sharp shells and body parts. Also, there may be snagged fishing hooks caught amongst the organisms. Small mussels can only be detected on fairly smooth surfaces, such as boat hulls, since tiny shells can hide in rough surfaces. Newly settled mussels feel like tiny individual bumps the size of large sand grains that are irregularly spaced. Similarly, larger juvenile and adult mussels are irregularly spaced and range in size from sunflower seeds to almonds. They are likely to be encountered in low numbers at water bodies where no infestation has been detected and preventative measures are in place. Thus, to distinguish newly settled mussels from other organisms or substances, feel for an anomalous object. Mussels can be tactilely distinguished from another organism or object by the way they rotate and stay attached when lightly pushed (see section 2.2. Identifying Quagga and Zebra Mussels).

Data sheets (Appendix IV) should be completed at the time the survey is being conducted, even if no mussels are found (zeros are very important!). Be sure to record the amount of linear feet that was surveyed for each surface survey. Take photographs of suspect organisms, but do not collect any samples (see section 4.4 Data Collection and Reporting).

Whether conducting visual and/or tactile surveys, it is critical to decontaminate gear, clothing, shoes and any part of yourself that contacted the water prior to leaving any monitoring site (see section *5. Decontamination*). Remember, it isn't just about the potential spread of mussels from site to site, but also the spread of other AIS (e.g., New Zealand mudsnails) that may be present at the water body.

### **Dive Surveys**

Underwater surveys, either snorkel or SCUBA (Self-Contained Underwater Breathing Apparatus), are useful for surveying quagga and zebra mussel populations because a diver can more closely assess mussel populations using both tactile and visual surveys at a variety of water depths. SCUBA surveys are particularly useful since divers can survey deeper water and remain underwater for long periods of time. While underwater surveys are desirable and successful volunteer dive monitoring programs exist, they require specific expertise, have increased liability issues associated with them and are often prohibited at many freshwater sites. For these reasons, we do not provide detailed information about these types of surveys. However, several government agencies include dive surveys in their mussel monitoring programs. Consult the administrators of the water body of interest or the DFG quagga/zebra regional staff (Appendix II) for more information.

### References

California Department of Water Resources Zebra/Quagga Mussels Surface Survey Protocol. <http://www.des.water.ca.gov/ docs/Zebra Quagga surface survey protocol. pdf>

Culver, Carolynn S. and Jenifer E. Dugan. Guide for Monitoring Recruitment of the Chinese Mitten Crab, *Eriocheir sinensis*. 2009. California Sea Grant College Program. 26pp.

U.S. Army Corp of Engineers. 2002. Zebra Mussel Information System: Monitoring of Settled Juveniles and Adults. *<http://el.erdc. usace.army.mil/zebra/zmis/>* 

### 4.2. Artificial Substrate Surveys

Surveys of artificial substrates are quite useful for monitoring sites where existing surfaces do not exist or are not visible or reachable. For example, artificial substrates can be deployed next to existing pipelines that are not visible from the surface. Likewise, the bottom of docks may not be reachable for thorough tactile surveys. Further, existing substrates may be absent down-current from busy boat ramps or discharge waterlines. Deploying artificial substrates in these areas enables monitoring of mussels at these otherwise inaccessible sites.

### Targeted Life Cycle Stage(s)

Artificial substrates are typically deployed for early- and post-detection monitoring of recently settled (post-larval) and juvenile mussels. This monitoring method takes advantage of the settlement behavior of early non-swimming stages where the mussels search for and settle on preferred substrates. Specific types of artificial substrates are used to facilitate settlement of mussels by providing surfaces that have characteristics the mussels prefer, and open space (habitat) for the mussels to attach to.

Certain artificial substrates are more effective for detecting mussels of specific sizes. Because young mussels (1/32"-1/8"; 1–3 mm) may first attach to filamentous substrates like aquatic weeds, artificial substrates with filamentous characteristics such as fibrous ropes, scrub brushes and scrub pads (Fig. 13) — are often used to monitor the smallest mussels. Larger juvenile mussels typically move to a hard substrate where they often stay for the remainder of their lives. Therefore, artificial hard substrates such as plastic (PVC/ABS) pipes and plates (flat pieces), cement and ceramic materials (Fig. 14) — are used to survey for larger iuvenile and adult mussels.

Artificial filamentous substrates in comparison to hard substrates not only target mussels of different sizes, they also require different processing methods and equipment. Because larger mussels can be detected easily without the aid of other equipment, we recommend using hard substrates at least initially when beginning your surveys. The lines that substrates are deployed on will provide some filamentous material for mussels to attach to, and they should be considered part of the monitoring effort. However, if you are interested in using more complex artificial filamentous substrates (i.e., scrub pads and brushes), please refer to Culver and Dugan (2009) for more details.



**Figure 13.** Artificial filamentous substrates used for monitoring young mussels and other invertebrates. A. Scrub brushes. B. Scrub pads (Tuffy<sup>®</sup>). Photographs by Carolynn Culver.



**Figure 14.** Artificial hard substrates used for monitoring mussels and other invertebrates. **A.** Plastic (PVC/ABS) pipe. **B.** Plastic (PVC/ABS) plates (flat pieces) — single substrate with vertical orientation on rope. **C.** Multiple plastic plates (flat pieces) — multiple substrates with horizontal orientation on rope. **D.** Cement block. **E.** Ceramic flower pot. **F.** Synthetic wood (Trex©). Photographs by Carolynn Culver.

### **Surveying Period and Frequency**

Artificial substrates typically are deployed a few weeks prior to the expected settlement period of an organism. For quagga and zebra mussels, artificial substrates should be deployed during months when the water temperature is at or above 48° F (9° C): the temperature at which quagga mussels begin reproducing. Depending on the conditions at the water body being monitored, this may mean monitoring for the entire year, or just during the warm season (likely May–October). If monitoring effort is limited, targeting the peak warm season (June–September) when conditions are optimal for mussel reproduction and growth will be best (Table 1).

Artificial substrates should be examined monthly, although data collected less frequently still can be helpful. Sampling every two weeks is only advised when water temperatures support optimal reproduction and growth. Daily and weekly sampling is not recommended as the frequent disturbance of the artificial substrates may minimize the ability of mussels to create a strong attachment. This can make it difficult to retrieve the artificial substrates without dislodging the mussels as the substrate(s) is pulled out of the water. Further, small mussels require natural biofilms — a layer of micro-organisms, including bacteria — for attachment. It takes a few days for these films to form naturally on the substrate after being submerged into a water body.

### Site Selection

Because there is a great deal of interplay between where you plan to monitor and how to assemble and deploy monitoring





**Figure 15.** Deployed artificial substrate survey lines. **A.** Line with a single substrate. **B.** Line with multiple substrates. \*Colored cable tie denotes the location of the surface of the water. This mark ensures that the substrates are attached at appropriate locations on the line and the line is deployed correctly to sample the intended water depth(s). Illustrations by Valerie Borel.

# equipment, please read through this entire subsection before heading out to choose sites.

Artificial substrates should be deployed in areas at high risk for mussel infestation (low light levels, slow water flow) (see section 3.4 Site Selection). When choosing sites, look for areas where it will be easy to attach survey lines to existing infrastructure or to install temporary structures without interfering with existing structures or their uses (see section Survey Lines). Be sure to consult the managing staff of the water body to determine average water depths, water clarity (light levels at various depths), water currents and potential attachment sites in the locations being considered for monitoring. Deploy artificial substrate at depths according to the specific parameters of the water body.

### **Substrate Selection**

Many different types of artificial substrates can be used for early detection and monitoring. While scientific testing of various substrates has been minimal in California, observations in the west and scientific testing elsewhere have found that some types of artificial substrates are better than others for monitoring quagga and zebra mussels. Examples of suitable artificial substrates include concrete, plastic (PVC, acrylic, ABS), polypropylene rope and other fibrous materials, fiberglass, wood, steel, and aluminum. Notably, while quagga and zebra mussels attach to many different types of artificial substrates, they don't attach to silicone and copper.

Choosing the type of artificial substrate to use is often a function of cost and the labor involved in preparing the artificial substrates for the survey line. Concrete blocks and ceramic flower pots have been used by some groups because they are inexpensive, easily obtainable and don't require modifications to attach to a survey line (Fig. 14D, E). These surfaces are attractive to settling mussels because they are porous and include many cracks, crevices, edges and corners. However, because these substrates are heavy and can chip or break, they are not always ideal. Instead, plastic (such as PVC or ABS) pipes and plates (flat pieces) are being used in agency-sponsored mussel monitoring programs (Fig. 14A, B, C). While these materials are more durable, lighter and not too expensive, they need to be cut into appropriate-sized pieces before they are attached to the monitoring line. We recommend roughing up plastic or other slick substrates with course sandpaper prior to attaching them to the monitoring line to increase the substrate texture. Currently, there are no data indicating whether one type of hard substrate (cement, plastic or ceramic) is more effective for monitoring guagga and zebra mussels in the Western United States. New scientific findings comparing various sampling techniques will help refine sampling protocols in the future.

Note: Post-detection monitoring programs require the use of similar types, numbers and sizes of artificial substrates to allow comparisons of the samples between and among sites. In contrast, early detection monitoring is only used for presence/absence, and thus any type, number and size of artificial substrates that are appropriate for mussel attachment may be used.

### **Survey Lines**

Each survey line consists of three parts: an attachment end, the artificial substrate(s) section, and a weighted end (Fig. 15A, B). Be sure that the piece of rope is long enough to accommodate all of these parts.

### The Attachment End

This section of the survey line spans from the attachment site to the surface of the water (Figs. 15, 16). Artificial substrate survey lines can be attached to either a: 1) permanent structure such as a dock, pier, walking bridge or overpass (Fig. 17A, B) or, 2) temporarily installed structure such as a mooring, buoy, sand anchor, stake, or rebar in the water or on the bank (Fig. 17C). The first option is typically easier and safer, but in high-use areas the readily noticeable lines may invite tampering or vandalism. Further, the lines could interfere with water activities. The second option allows better opportunities for camouflaging the sampling gear and for placing it in more remote locations outside



**Figure 16.** Deployed artificial horizontal substrate survey line with multiple substrates. Illustration by Valerie Borel.

high-use areas, but it requires installation of additional equipment. A temporary structure should be secured in the water body or far enough up on the bank to protect it from loss during extreme weather conditions, and in an area that will not interfere with other activities or pose a hazard.

### **The Substrate Section**

The substrate section spans from the surface of the water through the water column to just above the bottom of the survey line (Fig. 15). The number of artificial substrates will depend on the location being surveyed. If the target location is an existing structure (e.g., pipeline, dock) that occurs at a specific water depth, only one artificial substrate per vertical line will need to be deployed to that depth (Fig. 15A). However, if the target is a low light area in deep water, several artificial substrates should be spaced evenly along the vertical line to ensure a large portion of the water column is monitored (Fig. 15B). Remember, substrates should not be deployed near the surface of the water, unless the shallower portion of the water column is always shaded



Figure 17. Attachment sites for artificial substrate survey lines. A. Dock. B. Pier. C. Bank of a creek. Yellow arrows point to attachment location. Photographs by Carolynn Culver.

or in low light. We also recommend deploying sampling lines horizontally underneath docks to better monitor the large surface area of the underside of the dock (Fig. 16). In this case, several substrates should be used and spaced along the line at even intervals.

### The Weighted End

This contains a weight attached to the end of the line to keep the line straight when it is deployed in the water (Fig. 15A, B). More weight will be required for polypropylene line (because it floats) than for nylon rope (that absorbs water and sinks). Metal washers can be used when less weight is needed. When more weight is required, we recommend making weights by mixing and pouring concrete into large (16 oz/475 ml) plastic beverage cups with a small piece of chain, an eyebolt or looped rope from the top of the cup (Fig. 18). Alternatively, rocks, small pavers, bricks or other heavy materials can be attached to the end of the line to keep the line taut. Pieces of these materials are often provided at no cost at local home improvement stores. Note that an end weight is not required for horizontal lines, although some weight may be needed along the line to minimize tangling of the line with objects on the underside of the dock.

### Assembling the Survey Line

The following supplies will be needed to construct the artificial substrate survey line(s):

- Individual artificial substrate(s)
- Cable (zip) ties
- Rope (nylon, polypropylene)
- Weight(s) (metal washers, concrete weights)
- Wire cutters
- Duct tape

Cut a piece of rope the appropriate length for the survey site. Tape both ends with duct tape to minimize fraying. Measure the amount of rope needed for the attachment section and attach a colored cable tie at this location to represent where the survey line will meet the surface of the water. Using a color cable tie as a reference point for the surface of the water will ensure that the substrates are attached to the line at appropriate locations for the water depth(s) you intend to sample. It is only needed for lines that will be deployed vertically in the water column.

Then, measure the appropriate distance(s) and attach the artificial substrate(s) (Figs. 15, 16) so they will be at the desired water depth (i.e., shaded, low light areas). If many



**Figure 18.** Homemade concrete weights with material for attaching the weight to a line. **A.** Chain used as attachment material. **B.** Rope used as attachment material. Photographs by Carolynn Culver.

substrates are going to be used to survey a large portion of the water column, space the substrates out evenly (e.g., 5-10 ft apart; 1.5–3 m) along the line where they will be in the low light region of the water column. Substrates can be attached to the line with cable (zip) ties (Fig. 14B), or directly through the rope using knots to keep them in place (Fig. 14F). Even if cable ties are used, we recommend tying a knot above and below the substrate to provide additional filamentous substrate for settling mussels. Quagga mussels are often found in knots on monitoring lines, particularly polypropylene rope (D. Daft and C. Culver, pers. obs.). Substrates can be attached in a vertical or horizontal orientation to the line (Fig. 14B, C). Research indicates that mussels attach to substrates of both orientations, with preferences varying depending on the study. Importantly, the studies used different types of substrates and were conducted under different conditions. We suggest attaching the artificial substrates in a manner that is easiest for you.

Once the artificial substrates are secure, attach an appropriate sized weight to the end of the line using a cable (zip) tie, bowline or other secure knot. The line is now complete and is ready to be deployed at the survey site. It is a good idea to take a photograph of the assembled survey line for record keeping. PLEASE DO NOT DISTURB ECOLOGICAL MONITORING IN PROGRESS

UNIVERSITY OR GROUP NAME PERMIT NO.

CONTACT: (XXX) XXX-XXXX

**Figure 19.** Example of an identification label that is attached to each artificial substrate survey line.

### Deploying the Survey Line

Prior to deploying the survey line, be sure to make an identification label for each line (Fig. 19). This label should include your contact number and affiliation. We also recommend that the label includes a statement such as "Please Do Not Disturb, Ecological Monitoring In Progress." The identification labels should be durable so they are able to withstand local weather conditions. Often, this information is written in wax pencil on a small piece of flat white PVC material that is attached to the line by cable ties threaded through drilled holes. A cheaper alternative is to print out postcards with the appropriate information and then laminate the cards with plastic or clear contact paper, or place them in a small zip lock bag, to waterproof the cards. Punch a hole in the corner of each sealed label for attaching it to the line.

The following supplies will be needed to deploy your survey lines:

- Assembled artificial substrate lines
- Labels with contact information
- Cable (zip) ties
- Additional rope
- Wire cutters
- Attachment structures, if needed (floats, stakes, sand anchors)
- Duct tape

If needed, install the temporary attachment structure. Attach the survey line to the attachment site using a bowline or other secure knot. Once the line is secured to the attachment site, deploy the line into the water. Attach an identification label with your contact information to the deployed line at the point of attachment so that the label is out of the water.

### **Examining Artificial Substrates**

The following supplies will be needed to examine the artificial substrates:

- Bucket or rectangular plastic bin
- Data sheets
- Pencil
- Digital camera
- Wire cutters (to fix the line as needed)
- Cable ties (to fix the line or re-attach substrates as needed)

Artificial hard substrates are typically examined once a month during daylight hours, with activities suspended during hazardous conditions. Be sure to bring the supplies required for collecting and reporting your findings (see section *4.4. Data Collection and Reporting*) when you go to examine the survey lines.

To examine the artificial substrates, pull the weighted line up in a slow, continuous motion, avoiding hard surfaces (e.g., dock edge) to minimize loss of mussels that may not be firmly attached to the substrates. Watch and feel for mussels as the line is pulled from the water. Place the artificial substrates and line directly into a bucket or large bin as they come out of the water to contain any mussels that may become detached. If retrieving substrates from along the bank of the water body, take care to minimize dragging them along the river bank and through vegetation.

Once retrieved, closely examine the artificial substrates. Pay particular attention to corners, crevices, edges and seams. Be sure to look inside PVC tubes and flower pots. Once the artificial substrates have been examined, closely inspect the weight and the rope, especially around the areas where the substrates are attached and knots occur. Remember to look for other AIS, such as New Zealand mudsnails. When you have finished examining the artificial substrates, rope and weight, remove these items from the bucket, then examine the bottom of the bucket for mussels and other AIS that may have fallen off the substrates or lines. Photograph, but do not collect, any mussels or AIS you find. Take notes that describe what is shown in your photographs. What seems obvious or easily remembered at the survey site may not be so obvious to another person or even to you some months later. Also note on your data sheet exactly where the mussel was found; substrate type (e.g., rope, PVC pipe, PVC plate, etc.), where on substrate (e.g., inside pipe, bottom of plate, on rope knot) and water depth. Make sure your data sheets are complete, making an entry for every survey line examined even if no mussels were found (see section *4.4. Data Collection and Reporting*).

Once your survey is complete, re-deploy the survey line containing the artificial substrates. Make sure that the colored cable tie on the survey line is level with the surface of the water (if applicable). If it is not, adjust the line at the attachment site so it is at the appropriate level. Before leaving the site, decontaminate yourself and the gear using the procedures in section 5. *Decontamination*. Lastly, don't forget to report the survey results (see section 4.4. Data Collection and Reporting).

### References

Culver, Carolynn S. and Jenifer E. Dugan. Guide for Monitoring Recruitment of the Chinese Mitten Crab, *Eriocheir sinensis*. 2009. California Sea Grant College Program. 26pp.

Folino-Rorem, Nadine, James Stoeckel, Emily Thorn, and Laura Page. 2006. Effects of Artificial Filamentous Substrate on Zebra Mussel (*Dreissena polymorpha*) Settlement. *Biol. Invasions* 8: 89–96.

Kobak, Jaroslaw. 2001. Light, Gravity and Conspecifics as Cues to Site Selection and Attachment Behaviour of Juvenile and Adult *Dreissena polymorpha* (Pallas, 1771). *J. Moll. Stud.* 67: 183–189.

Marsden, J. Ellen and David M. Lansky. 2000. Substrate Selection by Settling Zebra Mussels, *Dreissena polymorpha*, Relative to Material, Texture, Orientation, and Sunlight. *Can. J. Zool* 78: 787–793.

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U.S. Army Corp of Engineers. 2002. Zebra Mussel Information System: Sampling Juveniles with Filamentous Substrates. <http://el.erdc.usace.army.mil/zebra/ zmis/zmishelp/sampling\_juveniles\_with\_ filamentous\_substrates.htm>

U.S. Army Corp of Engineers. 2002. Zebra Mussel Information System: Natural Solid Substrates. <http://el.erdc.usace.army. mil/zebra/zmis/zmishelp/natural\_solid\_ substrates.htm>



Figure 20. Plankton net. Photograph by Carolynn Culver.

### 4.3. Plankton Surveys

Plankton surveys sample for the presence of the veliger life stage of quagga and zebra mussels. As described in the *Background* section, veliger is a generic name for the free-swimming larval stage of the bivalve lifecycle.



**Figure 21.** Plankton sample viewed through microscope with zebra mussel veliger (arrow), copepods, diatoms and rotifers present. Photograph by Charles Ramcharan, Wisconsin Sea Grant.

Sampling for veligers can be useful if it is important to detect an infestation before adults have been found. However, it is a much more technical process than looking for juveniles and adults, and requires special equipment and expertise. Plankton nets (Fig. 20), filters and other materials are needed to collect the samples. The sample is examined under a microscope (Fig. 21) and/or chemically analyzed for the presence of quagga and zebra mussel DNA. These techniques are time-consuming, costly, and require technical expertise. If you are interested in conducting this type of survey, contact the local water body personnel and the DFG quagga/zebra regional staff (or your local fish and game authorities) to determine whether such surveys are desirable and where to obtain training and laboratory support.

### References

Crosier, Danielle and Daniel Molloy. 2002. Zebra Mussel Information System. U.S. Army Corps of Engineers. <http://el.erdc.usace. army.mil/zebra/zmis/>

Kelly, Kevin and Denise Hosler. 2008. Collecting Water Samples for *Dreissena* spp. Veliger Analysis. U.S. Bureau of Reclamation Technical Service Center, Denver, Co.

### 4.4. Data Collection and Reporting

An important part of early detection monitoring is the collection and reporting of the resulting data. In particular, it is critical to report findings even when nothing is detected — zeros are extremely important! Blank data sheets for recording your findings are provided in Appendix IV. These data sheets were developed for use by volunteer monitors and can be used to record data from multiple monitoring methods. If your monitoring is part of a larger coordinated effort, make sure all partners are utilizing the same data sheets and recording data in a consistent manner. Photocopies of the data sheets from this manual may be produced on regular or waterproof paper for use in the field. When generating your own data sheet, be aware that forms printed using an ink jet printer will smear if they become wet, even if on waterproof paper. Thus, if using an ink jet printer it is best to print the form and then make photocopies to be used in the field. This ink-smearing problem does not occur if the forms are printed using a laser printer or photocopier.

### **Data Collection**

Before leaving a site, record your findings for the site on the appropriate data sheet (Appendix IV). Fill out all information as completely as possible. If a GPS is not available, describe the site as accurately as possible using physical parameters that are not likely to change (street names, buoy number, distance from or location on permanent structures). Photographs of the site are also useful, and can aid you and others in returning to the same location. Take notes that help you and others remember exactly what is shown in the photographs. Remember to make an entry for every monitored site, even if no mussels were found. Numbering sites will help organize your records and make it easy to report your findings. If you find something you suspect to be a guagga or zebra mussel, take a picture and call the local DFG contact as soon as possible to arrange a site visit to examine the location where you found the suspect material. Do not collect any organisms or materials.

### **Reporting Your Findings**

After the surveys are completed, submit your data sheet(s) via email, mail, fax or telephone to the California DFG Regional Quagga/Zebra Mussel Staff (Appendix II) or your local fish and game authorities, as well as personnel of the local water body with whom you are coordinating your monitoring efforts. Portland State University would also appreciate receiving your information to include in their mussel monitoring database for the Western United States (see Appendix I *Monitoring*).

In California the DFG regional staff will forward results to state headquarters and others as necessary. DFG state headquarters is coordinating with the federal program (the 100th Meridian Initiative) to track quagga and zebra mussel occurrences nationwide.

# 5. Decontamination

To ensure that monitoring efforts do not spread AIS, it is essential to follow appropriate gear and personal decontamination protocols at the end of each early detection monitoring activity (Appendix IV). After monitoring for mussels at a site, be sure that you have decontaminated yourself and any gear or clothing (including shoes, socks, or booties) that contacted the water whether or not mussels have been found at that site. Decontamination procedures should be done at potentially infested sites in the same manner as at contaminated sites. This is particularly critical if you plan to immediately monitor another water body. Remember that even if water bodies do not contain guagga or zebra mussels, they could have other AIS that you should avoid spreading, such as New Zealand mudsnails. If at all possible, do not visit more than one water body on a single day. If visiting multiple water bodies in the same day or within a few days, have dedicated sets of gear, field clothes and shoes for each site.

Although not 100% effective, we recommend wiping yourself and your gear with a towel or scrubbing with a dry scrub brush before leaving a site to help dislodge unwanted AIS. In particular, wiping off your shoes — both tops and bottoms and under the laces — may minimize transport of AIS. However, due to the difficulty in visually inspecting for small AIS (e.g., New Zealand mudsnails), more effective procedures should also be used to minimize the likelihood of transferring live organisms among water bodies. Effective decontamination methods that kill mussels and other AIS include thoroughly drying all field clothes and shoes in a clothes dryer at high heat for 30 minutes. Exposing gear to full sun at low humidity for several days may also decontaminate clothes and gear, but it is important to make sure that every crevice dries completely for this method to be effective. To be safe, leave the item in the sun an additional 24 hours after it appears dry. For other gear, freezing the gear for 24 hours will eliminate most AIS. This method is most easily done by having a chest freezer specifically for decontamination. Chemical decontamination methods, as well as methods requiring exposure to very hot (>140° F) water, are available. Check with your DFG regional guagga/zebra mussel staff for their latest recommendations.

# 6. Appendices

# <u>Appendix I</u>

### Information and Resources About Quagga Mussels (*Dreissena bugensis*) and Zebra Mussels (*Dreissena polymorpha*)

### General Biology/Ecology/Distribution

### <u>Nationwide</u>

- General overview of the biology of both mussels. Developed by the U.S. Fish and Wildlife Service: *http://www.fws.gov/rubylake/quaggamussel.pdf*
- General information and a country-wide overview (with a map updated daily) for both mussels. Developed by US Geological Survey Nonindigenous Aquatic Species: http://nas.er.usgs.gov/taxgroup/mollusks/zebramussel/
- General overview by the 100th Meridian Project, a collaboration of several government agencies working on stopping the nationwide spread of invasive species: *http://www.100thmeridian.org/zebras.asp*

### <u>California</u>

- General information about quagga and zebra mussels and other invasive species in California, including reporting information from the California Department of Fish and Game: http://www.dfg.ca.gov/invasives/ quaggamussel/
- Current news about quagga and zebra mussels and their impact on statewide water supplies. Developed by the California Department of Water Resources: http://www.des.water.ca.gov/zmwatch/
- Local information may also be available by contacting your local water supplier.

### Preventing the Spread of Quagga and Zebra Mussels

### Inspections and Decontamination

Information on how to inspect vessels and prevent spreading aquatic nuisance species available at:

- California Department of Fish and Game: *http://www.dfg.ca.gov/invasives/ quaggamussel/*
- California Department of Water Resources: *http://www.des.water.ca.gov/zmwatch/what\_can\_i\_do/index.cfm*
- 100<sup>th</sup> Meridian Initiative: *http://www.100thmeridian.org/boats.asp*
- Protect Your Waters: http://www.protectyourwaters.net/prevention/ prevention\_generic.php and http://www.protectyourwaters.net/prevention/ user\_boaters.php

## Appendix I - continued

Monitoring Information & Trainings

- Monitoring information for California: *http://www.dfg.ca.gov/invasives/ quaggamussel/*. To participate in volunteer monitoring, please call the California Department of Fish Game's Quagga/Zebra Mussel Information Line at (866) 440-9530.
- Volunteer monitoring information for the Western United States developed by Portland State University: *http://www.clr.pdx.edu/projects/volunteer/zebra.php*

### Educational Resources

- California Department of Fish and Game, Don't Move a Mussel campaign materials: *http://www.dfg.ca.gov/invasives/quaggamussel/*
- California Department of Fish and Game's public education materials: *http://www.dfg.ca.gov/invasives/quaggamussel/docs/MoveMusselPoster.pd*f
- Pacific States Marine Fisheries Council Don't Move a Mussel DVD: *https://www.odva.state.or.us/OISC/mussel\_video.shtml*
- 100<sup>th</sup> Meridian Initiatives Zap the Zebra Campaign: *http://www.100thmeridian. org/ZTZ2007.asp*
- Protect Your Waters, Stop Aquatic Hitchhikers Campaign: *http://www.protectyourwaters.net/resources/#logo*
- Education about the problems caused by releasing unwanted pets and plants. Developed by Habitattitude: *http://www.habitattitude.net/*
- Exotic Aquatics on the Move Resource for K-12 education about aquatic invasive species. Developed by the National Sea Grant Network and Geographic Education Alliances: *http://www.iisgcp.org/edk-12/EXOTICSP/*

### General Management

- California Aquatic Invasive Species Management Plan: http://www.dfg.ca.gov/ invasives/plan
- California's Response to the Zebra/Quagga Mussel Invasion in the West: Recommendations of the California Science Advisory Panel: http://www.dfg. ca.gov/invasives/quaggamussel
- Zebra Mussel Rapid Response Plan: http://www.des.water.ca.gov/zmwatch/ zebra\_quagga/index.cfm

# Appendix II

# DFG Regional Quagga/Zebra Mussel Staff Contacts\*

### **Region 1 – Northern Region**

Counties: Del Norte, Humboldt, Lassen, Mendocino, Modoc, Shasta, Siskiyou, Tehama and Trinity 601 Locust Street Redding, CA 96001 L. Breck McAlexander LMCALEXANDER@dfg.ca.gov Office: (530) 225-2317 Fax: (530) 225-2381

### **Region 2 – North Central Region**

Counties: Alpine, Amador, Butte, Calaveras, Colusa, El Dorado, Glenn, Lake, Nevada, Placer, Plumas, Sacramento, San Joaquin, Sierra, Sutter, Yolo and Yuba 1701 Nimbus Road Rancho Cordova, CA 95670 Jason Roberts JDROBERTS@dfg.ca.gov Office: (916) 358-2895 Fax: (916) 358-2912

### **Region 3 – Bay Delta Region**

Counties: Alameda, Contra Costa, Marin, Napa, Sacramento, San Mateo, Santa Clara, Santa Cruz, San Francisco, San Joaquin, Solano, Sonoma and Yolo 4001 North Wilson Way Stockton, CA 95205 Catherine Mandella CMANDELLA@dfg.ca.gov Office: (209) 942-6107

Fax: (209) 946-6355

### **Region 4 – Central Region**

Counties: Fresno, Kern, Kings, Madera, Mariposa, Merced, Monterey, San Benito, San Luis Obispo, Stanislaus, Tulare and Tuolumne 1234 E. Shaw Avenue Fresno, CA 93710 Mark Watson MWATSON@dfg.ca.gov

Office: (559) 243-4017 x244 Fax: (559) 243-4022

### **Region 5 – South Coast Region**

Counties: Los Angeles, Orange, San Diego, Santa Barbara and Ventura 4949 Viewridge Avenue San Diego, CA 92123 Daniel Schrimsher DSCHRIMSHER@dfg.ca.gov Office: (858) 467-6926 Fax: (858) 467-4299

### Region 6 – Inland Deserts Region

Counties: Imperial, Inyo, Mono, Riverside and San Bernardino P.O. Box 2160 Blythe, CA 92226 David Vigil DVIGIL@dfg.ca.gov Office: (760) 922-4928 Fax: (760) 922-5638 Rick Francis RFRANCIS@dfg.ca.gov Cell: (760) 828-7582

Fax: (760) 922-5638

\*For updated information on individual contacts refer to *http://www.dfg.ca.gov/invasives/quagga-mussel* or contact your local Fish and Game office.

# Appendix III



# **Quick Reference Sheet**

**Generalized Monitoring Procedures** 



### **Recommended Gear**

Closed-toed shoes with aood tread Long-sleeved shirt and long pants Hat or visor Sunglasses Sunscreen

First aid kit Disinfectant hand wash Hand towel Buddy and/or contact person Cell phone Digital camera

Data sheet Pencils Notebook/paper (waterproof) \*Buckets or other bins \*Wire cutters \*Cable (zip) ties

\*Equipment needed for artificial substrate surveys to examine substrates and fix the lines as needed.

### **Existing Surface Surveys**

- 1. Conduct surveys every two weeks.
- 2. Visually and tactilely inspect all types of substrates and the shoreline.
- 3. Record size/amount/length of surface surveyed.
- 4. Record presence/absence of mussels.
- 5. Photograph mussels or other aquatic invasive species found. Do not collect.
- 6. Record detailed information about the photograph taken and the site found.
- 7. Decontaminate gear and yourself before leaving site.
- 8. Contact DFG at (866) 440-9530 to arrange for a site visit to examine possible mussels or other aquatic invasive species found.
- 9. Report findings from survey.

### **Artificial Substrate Surveys**

- 1. Conduct surveys once a month.
- 2. Pull survey line up out of water slowly and continuously.
- 3. Place substrates in bucket or large bin as they are retrieved from the water.
- 4. Visually and tactilely inspect the substrates for mussels and other aquatic invasive species (pay particular attention to corners, crevices, edges and seams).
- 5. Inspect rope and weight for invasive organisms.
- 6. Remove inspected substrates and line from bucket/bin and inspect the inside of the bucket/bin.
- 7. Record findings on data sheet.
- 8. Photograph mussels or other aquatic invasive species found. Do not collect.
- 9. Record detailed information about the photograph taken and the site found
- 10. Decontaminate gear and yourself before leaving site.
- 11. Re-deploy line, with colored cable tie at surface of the water.
- 12. Contact DFG at (866) 440-9530 to arrange for a site visit to examine possible mussels or other aquatic invasive species.
- 13. Report findings from survey.

# Appendix III - continued

**Decontamination** (required even if mussels are not found at site)

- 1. Decontaminate yourself and gear after each survey, and before leaving the survey site.
- 2. At site, wipe yourself and gear with a towel or scrub with a dry scrub brush. (Pay particular attention to the top and bottom and under the laces of shoes).
- 3. Remove footwear and place in a plastic container or bag (to avoid contaminating vehicle).
- 4. Once home, decontaminate shoes, clothes and gear by one of the following methods:
  - a. Thoroughly dry;
    - i. 30 minutes at high heat in clothes dryer is best;
    - ii. Expose to full sun and low humidity for several days.
  - b. Freeze for 24 hours.

### **Aquatic Invasive Species Identification Tools**





**Invasive New Zealand Mud Snail** (*Potamopyrgus antipodarum*). This aquatic invasive species has a spiral shell but no byssal threads. Typical adult snails are only 3–5mm in length, much smaller than adult mussels. Photograph by Jane and Michael Liu, Oregon Sea Grant.

Quagga/Zebra Mussel Hotline California (toll free) 866-440-9530

# Appendix IV

# **Quagga and Zebra Mussel Monitoring Data Sheets**

### Quagga and Zebra Mussel Volunteer Monitoring Surface Survey Data Sheet

Monitoring Information				
Name/Affiliation:				
Date:				
Name of Water Body:				
GPS Coordinates (Decimal Deg	rees, WSG84):			
Site Description:				
Contact Information (email or	telephone):			
Existing Surfaces and Obie	cts (Minimun	n surveving	distances provided	in parentheses)
Surface Type	*Linear Ft or # and Size Examined	**Number of Mussels Present	Photograph Number (if taken)	Notes and Observations
Boat Ramp Bottom (100 ft at marina; 200 ft if solitary ramp)				
Dock/Pilings (200 ft)				
Mooring/Buoy Line (200 ft)				
Anchor/Dock Cable (100 ft)				
Concrete/Wood Structures (100 ft)				
Shoreline (100 ft)				
Buoys				
Anchors				
Logs/Woody Debris				
Aquatic Vegetation Type:				
Other (specify)				
Other Observations				
Other Species Observed	Surface Type	Number Present	Photograph Number (if taken)	Notes and Observations
New Zealand Mud Snail				
Asian Clam				
Other Organisms Present (specify)				

\*Indicate number of linear feet examined for long surfaces. Indicate number and size of objects examined.

\*\*Estimate the number if > 30 mussels are present. Be sure to enter '0' if no mussels are detected.





Quagga Mussel Photo by Dave Brenner, Michigan Sea Grant Size: < 2" / 50 mm



New Zealand Mud Snail Photo by Jane and Michael Liu, Oregon Sea Grant Size: < 3/16" / < 5 mm (TINY!)



Asian Clam Photo by Noel Burkhead, The Nature Conservancy Size: < 1" / < 25 mm

Note: Only take photographs. Do not collect any organisms or materials.



# Quagga and Zebra Mussel Volunteer Monitoring **Artificial Substrate Survey Data Sheet**

Monitori	ng Inform	ation						
Name/Aff	iliation:			Contact Ini	formation (ema	il or phone):		Date:
Name of V	<b>Nater Body</b>			Site Descri	ption:			
GPS Coon	dinates (De	scimal Degre	ses, WSG84):					
Artificial	Substrate	es Survey						
Line	Substrate	Substrate	Substrate	Water	Number	Where	Photograph	Notes and Observations
Number <sup>1</sup>	Number <sup>2</sup>	Type <sup>3</sup>	Orientation (Vert/Horz)	Depth of Substrate	of Mussels Present <sup>4</sup>	Mussels were Located <sup>5</sup>	Number (if taken)	
Other Ob	oservation	S						
Other Sp	ecies Obser	ved	Line and Substrate #s	Number Present	Photograph # (if taken)	Notes and Obs	ervations	
New Zeala	nd Mud Snai	li						
Asian Clarr								
Other Org	anisms Prese	ent (specify)						

<sup>1</sup>Number your lines at each site. Keep a master list and map of locations and associated numbers.

<sup>2</sup>Substrate #1 should be the one closest to water surface and furthest from weight when more than one substrate is on the line.

<sup>3</sup>e.g., flat plate (plastic, wood, other), PVC pipe, cement block, other (specify).

<sup>4</sup>Estimate the number if > 30 mussels are present. Be sure to put '0' if no mussels are detected.

<sup>5</sup>e.g., top, bottom, side, inside of substrate; on line (rope) above/below substrate; on weight; other (specify).

# Quagga Mussel

Photo by Dave Brenner, Michigan Sea Grant

Size: < 2" / < 50 mm



Photo by Jane and Michael Liu, Oregon Sea Grant **New Zealand Mud Snail** 

Photo by Noel Burkhead, The Nature Conservancy

Asian Clam

Size: < 1" / < 25 mm



Size: < 3/16" / < 5 mm (TINY!)

Note: Only take photographs Do not collect any organisms or materials.