California Department of Fish and Wildlife
Aquatic Invasive Species Decontamination Protocol

Purpose

California Department of Fish and Wildlife (CDFW) is committed to protecting the state's diverse fish, wildlife, and plant resources, and the habitats upon which they depend. Aquatic Invasive Species (AIS) are non-native aquatic organisms that negatively impact our state's environment, economy, or human health. Preventing the spread of AIS and disease-causing pathogens in CDFW activities, CDFW-permitted activities, and all activities outside the purview of CDFW's authority protects the natural resources of the state.

Application of this protocol is intended to prevent the spread of AIS and pathogens by removing biological agents by physical or chemical means, henceforth referred to as 'decontamination.' This protocol was developed based on the best available science and identifies the decontamination methods empirically tested with the highest efficacy across the greatest number of AIS and pathogens of greatest concern in California.

Many AIS and pathogens are difficult, if not impossible, to see in the environment, and can be unknowingly transported in entrapped water and on equipment. Therefore, assume that any AIS or pathogen is or could be present, and decontaminate between watersheds (or locations within watersheds if site-specific concerns warrant), to prevent the spread of AIS.

All equipment, including but not limited to, wading, dive, and sampling equipment (e.g., water quality probes, nets, buckets, substrate samples, etc.); fishing gear; and watercraft, must be decontaminated using one or more of the protocols listed below. Use your best judgment and field sampling needs to select the method(s) appropriate for your location, equipment, and schedule.

This document is formatted to provide concise decontamination protocols (page 3-5), along with the supporting information for these recommendations (Appendix A. Summary of Decontamination Methods Considered and Their Efficacy by Species, and Appendix B. Aquatic Invasive Species and Pathogen Profiles). Any brand names used in this document do not represent endorsement by CDFW, but rather are referenced relative to their active ingredients and demonstrated efficacy in peer-reviewed publications for the purpose of AIS decontamination.

For more information about invasive species in California visit the California Department of Fish and Wildlife's Invasive Species Program webpage. For questions about AIS, implementing these protocols, or to report an invasive species sighting, email invasives@wildlife.ca.gov or call (866) 440-9530.

Best Practices for Working in Aquatic Systems

Along with decontamination, the following best practices should be implemented when practical to ensure activities minimize the potential to spread AIS:

- When working in flowing water begin upstream and work downstream to avoid transporting AIS upstream to uninfested areas.
- Only work in one waterbody per day and decontaminate your equipment at the end of the day.
- If you must work in multiple waterbodies, use separate equipment for each site and decontaminate it at the end of the day. Bag used equipment and keep separate from unused equipment to prevent cross-contamination.
- If you must work in multiple waterbodies in a single day and cannot use separate equipment, decontaminate it at the site you are leaving, prior to traveling to the next site. When feasible, conduct work in waters believed to be AIS-free first, and sites known or suspected to have AIS last.
- Felt soled boots are more difficult to decontaminate than rubber soled boots because they can retain moisture for more than a month. If possible, choose boots with rubber soles.
- Clean all equipment before decontaminating. Debris reduces the efficacy of all decontamination methods by sheltering organisms from exposure and/or neutralizing chemicals.

Equipment Decontamination for Aquatic Invasive Species and Pathogens

Not all decontamination methods are appropriate for all circumstances. Choose the decontamination method(s) appropriate for the equipment being decontaminated and site-specific considerations. When possible, opt for the method effective for the most AIS and pathogens as possible. If multiple methods must be used, perform them sequentially, and never mix chemicals.

Hot Water Immersion[1]: 50°C (122°F) for 30 minutes

CAUTION: *Wear appropriate personal protective equipment to avoid burns, such as heat resistant gloves and/or a splash apron. Hot water can damage heat-sensitive equipment. Do not use hot water to decontaminate equipment that cannot tolerate temperatures at or above 50°C (122°F).*

Step 1. Scrub equipment with a stiff bristled brush to remove all organisms, mud, debris, etc. Thoroughly brush small crevices such as boot laces, seams, net corners, etc. and rinse.

Step 2. Immerse equipment in 50°C (122°F) or hotter water for 30 minutes. Weigh equipment down if necessary. Maintain the water temperature at 50°C (122°F) for the duration of the 30-minute soak.

Drying[2]: 14°C (57°F) for 8 days OR 35°C (95°F) for 30 hours OR 70°C (158°F) for 15 minutes

Step 1. Scrub equipment with a stiff bristled brush to remove all organisms, mud, debris, etc. Thoroughly brush small crevices such as boot laces, seams, net corners, etc. and rinse.

Step 2. Allow equipment to dry completely. Once completely dry, maintain equipment completely dry at 14°C (57°F) for 8 days OR 35°C (95°F) for 30 hours OR 70°C (158°F) for 15 minutes.

NOTE: New Zealand mudsnails, quagga/zebra mussels, and other mollusks can take several days to lethally desiccate due to their protective shells. The post-dry clock begins following internal tissue desiccation of mollusks. Removing mollusks before drying reduces the time needed to achieve effective results for all other species.

---

[1] Tested effective for quagga/zebra mussels, didymo, chytrid fungus, whirling disease, sudden oak death, and white-nose syndrome. See Appendix A for more information.

[2] Tested effective for quagga/zebra mussels, New Zealand mudsnails, didymo, chytrid fungus, whirling disease, sudden oak death, and white-nose syndrome. See Appendix A for more information.

Virkon® S (21.41% potassium peroxymonosulfate + 1.5% sodium chloride) Immersion[3]: 2% solution for 20 minutes

CAUTION: *Virkon® S is a corrosive chemical known to degrade metals and materials over time. Avoid exposure to yourself and others, and prevent spills or contamination to the environment. Always read and follow all the product's label and Safety Data Sheet (SDS) procedures. Virkon® S must be mixed and used in a well-ventilated area, preferably outdoors. Wear appropriate personal protective equipment such as tightly fitting safety goggles (EN 166), chemical resistant gloves, and a splash apron to avoid contact with eyes, skin, and clothing. This chemical is known to cause serious damage to eyes if exposed; when working in the field bring a travel eye wash station.*

Step 1. Scrub equipment with a stiff bristled brush to remove all organisms, mud, debris, etc. Thoroughly brush small crevices such as boot laces, seams, net corners, etc. and rinse.

Step 2. Following the product label instructions mix a 2% solution.

Step 3. Immerse equipment in the 2% solution and soak for 20 minutes. Weigh equipment down if necessary.

Step 4. Rinse equipment and dispose of solution and rinse water in a municipal sewer system. Never dispose of solution or rinse water where it can drain into a waterbody.

[3] Tested effective for larval quagga mussels, New Zealand mudsnails, didymo, chytrid fungus, and ranavirus, and white-nose syndrome. See Appendix A for more information.

Watercraft Decontamination for Dreissenid Mussels

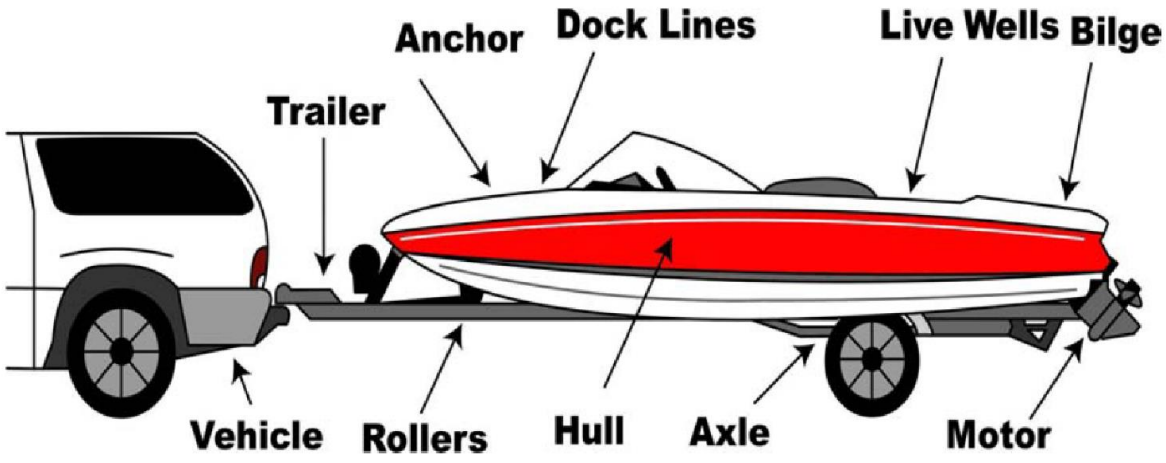
The following method is only known to be effective for decontaminating watercraft for adult dreissenid (quagga and zebra) mussels. It is not empirically known to be effective for any other species, however it is likely to be effective at mechanically removing all species of AIS and pathogens, and therefore is recommended for all watercraft to reduce the potential of transferring AIS and pathogens between locations.

Hot Water Spray[4]: 60°C (140°F) for a minimum of 10 seconds

Step 1. Clean all plants, debris, and mud from the watercraft, anchor and rope, trailer, and equipment. Dispose of all material in the trash.

Step 2. Drain all water from the watercraft, trailer, and equipment, including motor, motor cooling system, live wells, bilges, and lower end unit.

Step 3. Apply high-pressure, hot water 60°C (140°F) for a minimum of 10 seconds to all exterior and interior parts that have come into contact with the waterbody. Water temperatures can fluctuate considerably at the nozzle as the pressure washer cycles, and as the water moves through a watercraft, so it may be necessary to measure the temperature of the water at the output of the nozzle and as water drains from the watercraft to ensure an application temperature of 60°C (140°F) is achieved.



[4] Tested effective for adult quagga/zebra mussels. See Appendix A for more information.

References

Alonso, A., and P. Castro-Diez. 2012. Tolerance to air exposure of the New Zealand Mudsnail *Potamopyrgus antipodarum* (*Hydrobiidae*, Mollusca) as a prerequisite to survival in overland translocations. NeoBiota 14:67-74.

Anderson, L.G., A.M. Dunn, P.J. Rosewarne, and P.D. Stebbing. 2015. Invaders in hot water: a simple decontamination method to prevent the accidental spread of aquatic invasive non-native species. Biological Invasions 17:2287-2297.

Beyer, J., P. Moy, and B. De Stasio. 2011. Acute upper thermal limits of three aquatic invasive invertebrates: hot water treatment to prevent upstream transport of invasive species. Environmental Management 47:67-76.

Britton, D.K., and S. Dingman. 2011. Use of quaternary ammonium to control the spread of aquatic invasive species by wildland fire equipment. Aquatic Invasions 6(2):169-173.

Bryan, L.K., C.A. Baldwin, M.J. Gray, and D.L. Miller. 2009. Efficacy of select disinfectants at inactivating Ranavirus. Diseases of Aquatic Organisms 84:89-94.

Comeau, S., S. Rainville, W. Baldwin, E. Austin, S. Gerstenberger, C. Cross, and W.H. Wong. 2011. Susceptibility of quagga mussels (*Dreissena rostriformis bugensis*) to hot-water sprays as a means of watercraft decontamination. Biofouling 27(3):267-274.

De Stasio, B.T., C.N. Acy, K.E. Frankel, G.M. Fritz, and S.D. Lawhun. 2019. Tests of disinfection methods for invasive snails and zooplankton: effects of treatment methods and contaminated materials. Lake and Reservoir Management 35(2):156-166.

Dwyer, W.P., B.L. Kerans, and M.M. Gangloff. 2003. Effect of acute exposure to chlorine, copper sulfate, and heat on survival of New Zealand mud snails. Intermountain Journal of Sciences 9(2/3):53-58.

Gold, K.K., P.D. Reed, D.A. Bemis, D.L. Miller, M.J. Gray, and M.J. Souza. 2013. Efficacy of common disinfectants and terbinafine in inactivating the growth of *Batrachochytrium dendrobatidis* in culture. Diseases of Aquatic Organisms 107:77-81.

Hedrick, R.P., T.S. McDowell, K. Mukkatira, E. MacConnell, and B. Petri. 2008. Effects of freezing, drying, ultraviolet irradiation, chlorine, and quaternary ammonium treatments on the infectivity of myxospores of *Myxobolus cerebralis* for *Tubifex tubifex*. Journal of Aquatic Animal Health 20:116-125.

Hosea, R.C., and B. Finlayson. 2005. Controlling the spread of New Zealand mud snails on wading gear. California Department of Fish and Game Office of Spill Prevention and Response Administrative Report 2005-02.

Johnson, M.L., L. Berger, L. Philips, and R. Speare. 2003. Fungicidal effects of chemical disinfectants, UV light, desiccation and heat on the amphibian chytrid *Batrachochytrium dendrobatidis*. Diseases of Aquatic Organisms 57:255-260.

Kilroy, C., A. Lagerstedt, A. Davey, and K. Robinson. 2006. Studies on the survivability of the invasive diatom *Didymosphenia geminata* under a range of environmental and chemical conditions. Prepared for Biosecurity New Zealand. NIWA Client Report: CHC2006-116.

McMahon R.F, T.A. Ussery, and M. Clarke. 1993. Use of emersion as a zebra mussel control method. USACE (US Army Corps of Engineers) Zebra Mussel Research Program.

Moffitt, C.M., A. Barenberg, K.A. Stockton, and B.J. Watten. 2015. Efficacy of two approaches for disinfecting surfaces and water infested with quagga mussel veligers. Pages 467-477 in W.H. Wong and S.L. Gerstenberger, editors. Biology and management of invasive quagga and zebra mussels in the Western United States. CRC Press, Boca Raton, Florida.

Morse, J.T. 2009. Assessing the effects of application time and temperature on the efficacy of hot-water sprays to mitigate fouling by *Dreissena polymorpha* (zebra mussels Pallas). Biofouling 25(7):605-610.

Richards, D.C., P. O'Connell, and D.C. Shinn. 2004. Simple control method to limit the spread of the New Zealand mudsnail *Potamopyrgus antipodarum*. North American Journal of Fisheries Management 24:114-117.

Rzadkowska, M., M.C. Allender, M. O'Dell, and C. Maddox. 2016. Evaluation of common disinfectants effective against *Ophidiomyces ophiodiicola*, the causative agent of snake fungal disease. Journal of Wildlife Diseases 52(3):759-762.

Schisler, G.J., N.K. Vieira, P.G. Walker. 2008. Application of household disinfectants to control New Zealand Mudsnails. North American Journal of Fisheries Management. 28:1172-1176.

Schweigkofler, W., K. Kosta, V. Huffman, S. Sharma, K. Suslow, and S. Ghosh. 2014. Steaming inactivates *Phytophthora ramorum*, causal agent of Sudden Oak Death and ramorum blight, from infested nursery soils in California. Plant Health Progress doi:10.1094/PHP-RS-13-0111.

Shelley, V., S. Kaiser, E. Shelley, T. Williams, M. Kramer, K. Haman, K. Keel, and H.A. Barton. 2013. Evaluation of strategies for the decontamination of equipment for *Geomyces destructans*, the causative agent of White-Nose Syndrome (WNS). Journal of Cave and Karst Studies 75(1):1-10.

Stockton, K.A., and C.M. Moffitt. 2013. Disinfection of three wading boot surfaces infested with New Zealand mudsnails. North American Journal of Fisheries Management 33(3):529-538.

Stockton-Fiti, K.A., and C.M. Moffitt. 2017. Safety and efficacy of Virkon® aquatic as a control tool for invasive mollusks in aquaculture. Aquaculture 480:71-76.

Stout, J.B., B.W. Avila, and E.R. Fetherman. 2016. Efficacy of commercially available quaternary ammonium compounds for controlling New Zealand mudsnails, *Potamopyrgus antipodarum*. North American Journal of Fisheries Management 36:277-284.

Virkon® S; EPA RN: 39967-137; Material Number 57818065; LANXESS Corporation: Pittsburgh, PA. http://virkon.us/wp-content/uploads/sites/15/2017/11/VirkonTM-S-USA.pdf (accessed august 11, 2022).

Wagner, E.J., M. Smith, R. Arndt, and D.W. Roberts. 2003. Physical and chemical effects on viability of the *Myxobolus cerebralis triactinomyxon*. Diseases of Aquatic Organisms 53(2):133-142.

Appendix A. Summary of Decontamination Methods Considered and Their Efficacy by Species

CDFW's recommendations for the safest and most effective decontamination options are immersion in hot water, drying, or immersion in a 2% solution of Virkon® S for 20 minutes. These methods are relatively safe (in comparison to bleach and quaternary ammonium compounds) to implement and demonstrated to be effective at killing the most species of AIS and pathogens (Table 1). Hot water and drying have the advantage of being environmentally benign.

Other decontamination methods have been demonstrated to be effective for a variety of species (Table 1) but are not recommended for general use because they are not demonstrated effective across a wide range of species of concern or require added handling precautions that make them poor choices for use in the field. In addition, quaternary ammonium compound formulations vary and have tended to change, so it is difficult to assess which ingredient, or combination of ingredients, are responsible for the product's efficacy. As a result, we are unable to make a science-based recommendation on the efficacy of quaternary ammonium compounds.

Table 1. Summary of research demonstrating the efficacy of different decontamination methods by species. "EFFECTIVE" indicates that the method, applied as described in the cited publication, resulted in 100% mortality of the species. "NOT EFFECTIVE" indicates it resulted in less than 100% mortality of the species. "No Data" means no compelling published efficacy information was found. "Recommendation" is the treatment (temperature/duration or concentration) effective across all species for which efficacy data was available. Not all chemicals are recommended due to their efficacy and safety considerations, but are included to summarize all decontamination methods considered. All temperatures are provided in °C only.

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
Hot Water (Immersion)	Anderson et al. 2015 Adult zebra 45°C/15 min EFFECTIVE Beyer et al. 2011 Adult quagga and zebra 43°C/5 min EFFECTIVE	No Data	Kilroy et al. 2006 40°C/20 min; 60°C/1 min EFFECTIVE	No Data	Johnson et al. 2003 100°C/1 min; 60°C/5 min; 47°C/30 min; 37°C/4 hr EFFECTIVE	No Data	Wagner et al. 2003 75°/5 min EFFECTIVE	Schweigkofler et al. 2014 50°C/30 min EFFECTIVE	Shelley et al. 2013 50°C/20 min EFFECTIVE	50°C/30 min

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
Hot Water Spray (Specific to watercraft)	Comeau et al. 2011 Adult quagga 60°C/5 sec EFFECTIVE Morse 2009 Adult zebra 60°C/10 sec EFFECTIVE	No Data	No Data	No Data	No Data	No Data	No Data	No Data	No Data	60°C/10 sec for watercraft for quagga and zebra mussels only
Freezing	McMahon et al. 1993 Adult zebra -1.5°C/15 hr EFFECTIVE	Richards et al. 2004 -3°C/2 hr EFFECTIVE	Kilroy et al. 2006 -2°C/4 hr EFFECTIVE	No Data	No Data	No Data	Hendrick et al. 2008 -20°C/7 day EFFECTIVE Wagner et al. 2003 -20°C/1 hr EFFECTIVE	No Data	No Data	-20°C/1 hr Not recommended based on lack of efficacy data across taxa

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
Drying (Ambient temperature or heat)	Anderson et al. 2015 Adult zebra 14 ± 1°C/~8 day EFFECTIVE McMahon et al. 1993 Adult zebra 35°C/<5% relative humidity/30 hr; 35°C/>95% relative humidity/14 hr EFFECTIVE	Alonso and Castro-Diez 2012 15°C/53 hr EFFECTIVE Dwyer et al. 2003 45°C/60 sec EFFECTIVE Richards et al. 2004 40°C/1 hr EFFECTIVE	Kilroy et al. 2006 28°C/3 day EFFECTIVE	No Data	Johnson et al. 2003 32°C/4 day; 37°C/4 hr; 47°C/30 min; 60°C/5 min EFFECTIVE	No Data	Hedrick et al. 2008 20°C/2 months; 18-42°C/105 min sun; 22°C/18.5 hr indoors EFFECTIVE Wagner et al. 2003 1 hr EFFECTIVE	Schweigkofler et al. 2014 60°C/30 min EFFECTIVE	Shelley et al. 2013 70°C/15 min EFFECTIVE	14°C/8 day; 35°C/30 hr; 70°C/15 min
Bleach	No Data	De Stasio et al. 2019 400 mg/L NOT EFFECTIVE Dwyer et al. 2003 3000 mg/L/90 sec NOT EFFECTIVE Hosea and Finlayson 2005 5% (3000 mg/L sodium hypochlorite) /5°C & 15°C/5 min NOT EFFECTIVE	Kilroy et al. 2006 2%/1 min EFFECTIVE	Rzadkowska et al. 2016 3%/2 min EFFECTIVE	Gold et al. 2013 3%/1 min EFFECTIVE Johnson et al. 2003 0.2-0.01%/10 min EFFECTIVE	Bryan et al. 2009 3%/1 min EFFECTIVE	Hedrick et al. 2008 2500mg/L/15 min EFFECTIVE Wagner et al. 2003 0.25% (130ppm)/10 min EFFECTIVE	No Data	Shelley et al. 2013 0.6%/10 min EFFECTIVE	Not recommended based on safety considerations

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
Quaternary Ammonium Compounds (Various products and formulations)	Britton and Dingman 2011 Veliger Quagga "Sparquat 256" 3%/5 min EFFECTIVE	De Stasio et al. 2019 "Formula 409"/20 min EFFECTIVE Hosea and Finlayson 2005 "Formula 409" (undiluted and 50%)/5°C/5 min EFFECTIVE Schisler et al. 2008 "Formula 409" (undiluted)/10 min; 3.1+% Sparquat 256 /10 min; EFFECTIVE Stout et al. 2015 "GS 256" (undiluted)/10 min; "Super HDQ" (undiluted)/5 min; EFFECTIVE	Kilroy et al. 2006 "303 Clearall" 1.5%/1 min EFFECTIVE	Rzadkowska et al. 2016 "Process NPD" 0.4%/10 min; "409"/10 min EFFECTIVE	Johnson et al. 2003 "Path-X" diluted 1 X 10^{-3}/30 sec; "quaternary ammonium compound 128" 7.68% DDAC 1 x 10^{-3}/30sec; EFFECTIVE	No Data	Hedrick et al. 2008 ADBAC ("409" 1500mg/L and "Scott Fly Rod" 1000mg/L) 0.3% (1500mg/L)/10 min EFFECTIVE	No Data	Shelley et al. 2013 "Formula 409"/10 min; "Lysol IC" (1:128)/10 min EFFECTIVE	Not recommended based on safety considerations and uncertainty of efficacy across various product formulations

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
Virkon® (Immersion)	Moffitt et al. 2015 "Virkon Aquatic" Veliger quagga 5g/L/20°C/10 min EFFECTIVE	De Stasio et al. 2019 "Virkon Aquatic" 2%/20 min EFFECTIVE Stockton and Moffitt 2013 "Virkon Aquatic" 2%/15°C/30 min EFFECTIVE Stockton-Fiti and Moffitt 2017 Adult Quagga "Virkon Aquatic" 10 g/L/8°C; 15°C; 22°C/20 min EFFECTIVE	Kilroy et al. 2006 "Virkon S" 1%/30 sec EFFECTIVE	No Data	Johnson et al. 2003 "Virkon" 1mg/ml^{-1}/20 sec EFFECTIVE Gold et al. 2013 "Virkon S" 1%/1 min EFFECTIVE	Bryan et al. 2009 "Virkon S" 1%/1 min EFFECTIVE	No Data	No Data	Shelley et al. 2013 "Virkon" 1% NOT EFFECTIVE	2%/20 min

Decontamination Method by Species	**Quagga** (*Dreissena rostriformis bugensis*) and **Zebra Mussel** (*Dreissena polymorpha*)	**New Zealand Mudsnail** *Potamopyrgus antipodarum*	***Didymo*** *Didymosphenia geminata*	***Snake Fungal Disease*** *Ophidiomyces ophidiicola*	***Chytrid Fungus*** *Batrachochytrium dendrobatidis (Bd)*	***Ranavirus***	***Whirling Disease*** *Myxobolus cerebralis*	***Sudden Oak Death*** *Phytophthora ramorum*	***White-Nose Syndrome*** *Pseudogymnoascus destructans*	***Recommendation***
70% Ethanol	No Data	No Data	Kilroy et al. 2006 70%/10 min EFFECTIVE	Rzadkowska et al. 2016 70%/2 min EFFECTIVE	Johnson et al. 2003 70%/20 sec EFFECTIVE	No Data	No Data	No Data	Shelley et al. 2013 70%/10 min NOT EFFECTIVE	Not recommended based on lack of efficacy data across taxa

Appendix B. Aquatic Invasive Species and Pathogen Profiles

Quagga Mussel (*Dreissena rostriformis bugensis*)

Identification

Quagga mussels are small freshwater mussels of variable color, dark and light alternating stripes, but they may also be solid cream, brown, or black. Quagga mussels are very similar in appearance to zebra mussels (*Dreissena polymorpha*). Quagga mussels attach to surfaces with byssal threads and range from the size of a grain of sand (newly settled juveniles) to the size of a fingernail. Larvae are microscopic and free-floating in the water. Adults can survive over 30 days out of water in cool, humid conditions. Adults and larvae can be unknowingly moved between waters attached to watercraft and entrapped in livewells, bilges, and motors.

Impacts

Quagga mussels multiply quickly and out-compete other species for food and space. Their presence can alter food webs and environments, which negatively affects native and game fish species. Quagga mussels attach to hard and soft surfaces, impacting water delivery systems, hydroelectric facilities, agriculture, recreational boating, and fishing.

Habitat

Freshwater ranging from surface depth to more than 37 meters (400 ft). Veliger larvae are free-floating in the water column. Juveniles and adults attach to both hard and soft surfaces.

Distribution

A map of known quagga and zebra mussel distribution in California.

Zebra Mussel (*Dreissena polymorpha*)

Identification

Zebra mussels are small freshwater mussels of variable color, usually dark and light alternating stripes, but they may also be solid cream, brown, or black. Zebra mussels are very similar in appearance to quagga mussels (*Dreissena rostriformis bugensis*). Zebra mussels attach to surfaces with byssal threads and range from the size of a grain of sand (newly settled juveniles) to the size of a fingernail. Larvae are microscopic and free-floating in the water. Adults can survive over 30 days out of water in cool, humid conditions. Adults and larvae can be unknowingly moved between waters attached to watercraft and entrapped in livewells, bilges, and motors.

Impacts

Zebra mussels multiply quickly and out-compete other species for food and space. Their presence can alter food webs and environments, negatively affecting native and game fish species. Zebra mussels attach to hard and soft surfaces, impacting water delivery systems, hydroelectric facilities, agriculture, recreational boating, and fishing.

Habitat

Freshwater, from surface depth to more than 55 meters (180 feet) deep. Veliger larvae are free-floating in the water column. Juveniles and adults attach to hard surfaces.

Distribution

A map of known quagga and zebra mussel distribution in California.

New Zealand Mudsnail (*Potamopyrgus antipodarum*)

Identification

New Zealand mudsnail (NZMS) average 3 mm in length, but young snails may be as small as a grain of sand. The shell of NZMS is dark to light brown, elongated, and typically has 5-6 whorls (spirals). Whorls are dextral (spiral upwards to the right). Expert identification is necessary to confirm the identification of NZMS.

Impacts

New Zealand mudsnails are ovoviviparous and parthenogenic, meaning that they bear live young, and these young are clonal females that reproduce asexually. Population density has reached as high as 750,000 individuals per square meter. NZMS out-compete and replace native invertebrates that are the preferred food source of many fish species, but NZMS are themselves an inferior food source.

Habitat

New Zealand mudsnails are found on a wide variety of substrates and vegetation in fresh and brackish lakes, rivers, streams, and estuaries.

Distribution

A map of known NZMS locations in California.

Didymo (*Didymosphenia geminata*)

Identification

Didymo is a large diatomaceous alga that can create large, harmful blooms. Didymo attaches to submerged stones, aquatic plants, and other submerged materials. Clumps can be brown to dull yellowish to white in color and feel like wet cotton wool. Didymo can spread to new areas with the transport of a single microscopic cell. Because this alga is microscopic until large colonies form, didymo may not be visible until it is well established in the environment.

Impacts

Didymo's impacts are minimal if present in low numbers. However, the massive blooms usually seen can reduce benthic habitat for fish, invertebrates, plants, and other algae, and are thought to alter invertebrate communities. Decomposition of large mats may deplete dissolved oxygen. Didymo blooms also significantly reduce the aesthetics of affected waters.

Habitat

Didymo is found in freshwater rivers, streams, and lakes. It requires high light levels and favors consistent flow conditions. Didymo has been found from the surface to depths of 2 meters in ideal light conditions.

Distribution

The native range of didymo seems to have been broadly throughout the temperate Northern Hemisphere. However, until recently, aggressive blooms were not recorded. In California, didymo has been reported in the American River.

Snake Fungal Disease (*Ophidiomyces ophidiicola*)

Identification

Snake fungal disease is caused by the keratinophilic fungus *Ophidiomyces ophidiicola*. Infected snakes often show symptoms of skin lesions, skin swelling, and pustules; all of which affect the body and often the head. Mortality is variable based on snake species. The disease affects both terrestrial and aquatic snake species and is a soil borne fungus.

Impacts

Affects the skin, deep muscle tissue, and bone. Distribution of this disease was previously contained to the East Coast. Northern California has had several occurrences in different locations, indicating the disease has been transferred to multiple populations of both terrestrial and aquatic snakes. This disease could potentially wipe out native species, most of which are already considered at risk.

Habitat

Ophidiomyces ophidiicola is found in soil and water. Spores disperse through both.

Distribution

Current known distribution is Europe, Australia, and the United States. Most states east of the Mississippi River.

Chytrid Fungus (*Batrachochytrium dendrobatidis*; Bd)

Identification

Chytridiomycosis disease in frogs is caused by the fungus *Batrachochytrium dendrobatidis (Bd)*. Infected frogs may exhibit no symptoms, or may exhibit lethargy, partially closed eyes, and/or sloughing skin. However, these symptoms may also have other causes. Laboratory testing is necessary to positively identify chytrid fungus.

A related pathogen, *Batrachochytrium salamandrivorans* (*Bsal*), first identified in fire salamanders (*Salamandra salamandra*) in the Netherlands, has not yet been detected in the United States. However, *Bsal* is a serious potential threat to amphibian populations, particularly salamanders, in the western hemisphere.

Impacts

Chytrid fungus infects the keratinized parts of amphibians, including the skin of adults and the mouthparts of larvae, ultimately leading to death. Chytridiomycosis is believed to be a leading cause of the decline of native amphibian populations all over the world and has been responsible for the extinction of multiple species. Vulnerability varies based on species and even population, with some populations experiencing 100% mortality, while others are less affected. The invasive American bullfrog, *Lithobates catesbeianus,* has been identified as a carrier of chytrid fungus.

Habitat

Chytrid fungus is found in soil and water. The spores disperse through water.

Distribution

Nearly worldwide.

Ranaviruses

Identification

Ranaviruses are DNA-based viruses belonging to multiple taxonomic genera that infect amphibians, reptiles, and fish. Infected amphibians often present with skin lesions, especially at the vent and base of the hind limbs; lethargy; erratic swimming; and accumulation of fluid under the skin (edema). Turtles infected with a Ranavirus usually present with respiratory difficulty, lethargy, swelling, hemorrhages, and skin ulcers. Infected turtles may also show white to yellow plaques in the mouth, and lesions on the bottoms of their feet. Infected fish may present with loss of buoyancy, erratic swimming, swollen gills, hemorrhaging, and an overinflated swim bladder. A massive die-off of a single or multiple species may be indicative of Ranaviruses.

Impacts

Ranaviruses have been identified in at least 175 species among 52 families of ectothermic vertebrates. Die-offs can be massive, comprising thousands of individuals. Typically, in western states, die-offs are limited to a single species due to limited amphibian diversity.

Habitat

Wherever host species are found. Most at risk are mole salamanders (*Ambystoma* spp.), true frogs (*Lithobates* spp. and *Rana* spp.), and box turtles (*Terrapene* spp.).

Distribution

Likely present in every continental U.S. state. Ranaviruses have been found on all continents except Antarctica.

Whirling Disease (*Myxobolus cerebralis*)

Identification

Whirling disease in salmon and trout is caused by the parasite Myxobolus cerebralis. The parasite has a two-host life cycle, relying on the aquatic worm Tubifex tubifex and salmonids as hosts. Infected fish typically present with a darkened tail and skeletal deformities that result in a bent tail and characteristic whirling swimming pattern.

Impacts

Although infection by *Myxobolus cerebralis* is not itself fatal, the inability of infected fish to swim normally prevents them from feeding effectively, which makes them easy prey for predators. Parasitic spores of *Myxobolus cerebralis* can persist in sediment for 20 years.

Habitat

Myxobolus cerebralis requires salmonids and the worm *Tubifex tubifex* in order to complete its lifecycle, both of which are common in freshwater habitats.

Distribution

Myxobolus cerebralis is native to Europe and is found in most U.S. states including California, Oregon, Washington, and Nevada.

Sudden Oak Death Syndrome (*Phytophthora ramorum*)

Identification

Sudden oak death is caused by the water mold Phytophthora ramorum. This fungus infects a variety of trees and shrubs; foliar host species are not typically at risk of mortality and exhibit different symptoms than terminal hosts. In California, the most common foliar host is the California bay laurel (Umbellularia californica). Infected foliar hosts typically show leaf spots. Tanoak (Notholithocarpus densiflorus) is the most susceptible terminal host, but coast live oak, California black oak, Shreve oak, and canyon live oak are also terminal hosts. Terminal hosts infected by Phytophthora ramorum typically exhibit weeping trunk cankers and, later, browning of leaves throughout the tree. The symptoms of Phytophthora ramorum infection in both foliar and terminal hosts are very similar to symptoms of other diseases or insect infestation, so positive identification requires laboratory analysis of a sample.

Impacts

Terminal hosts infected by *Phytophthora ramorum* typically die within 2 years of infection. Sudden oak death has caused the death of millions of tanoaks and oak species. Infected trees are also more susceptible to attack by insect pests.

Habitat

Phytophthora ramorum requires cool, moist habitats, where its host species are present. Distribution of *Phytophthora ramorum* in California can best be predicted by presence and distribution of California bay laurel.

Distribution

Found in Europe and North America, including California.

White-Nose Syndrome (WNS; *Pseudogymnoascus destructans*)

Identification

White-nose syndrome in bats is caused by the fungus *Pseudogymnoascus destructans*. Infected bats often exhibit white fungal growth on the nose and wings. Large die-offs of bats are often another sign of WNS. However, laboratory identification is necessary to conclusively identify WNS.

Impacts

Pseudogymnoascus destructans causes physiological changes in infected bats that eventually lead to death. Entire colonies can be killed by the fungus. In the eastern U.S., *Pseudogymnoascus destructans* is estimated to have killed more than 6 million bats.

Habitat

Pseudogymnoascus destructans requires its host species and grows during winter hibernation of the bats. So far in North America, seven species have been affected, two of which (little brown bats and big brown bats) are found in California. An additional 12 species of California bats are thought to be at risk.

Distribution

White-nose syndrome occurrence is predominantly in the eastern United States but was detected in Washington in 2016. In July 2019 surveillance in California suggested the fungus that causes WNS is now present in California. At this time there is no indication the disease itself has taken hold in the local bat populations, however surveillance results like these have preceded WNS occurrence elsewhere in the country by one to a few years.