RECLAMATION
Managing Water in the West

U.S. Department of the Interior
Bureau of Reclamation

Research Update

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Bottom Line

Reclamation uses both microscopy and polymerase chain reaction (PCR) for the early detection of dreissenid mussels. PCR results are variable, and it is possible to get a negative result by microscopy and a positive result by PCR on the same sample. This research increases understanding of the factors that lead to variable test results.

Better, Faster, Cheaper

Invasive mussels are aggressive biofoulers that threaten water delivery and hydropower reliability. Using microscopy and PCR for sample analysis can increase the chance of mussel detection. Early detection will allow water managers additional time to prepare for the impacts of a new infestation.

Detecting Free-Floating Quagga Mussel DNA

Examining factors that impact successful detection of quagga mussel DNA

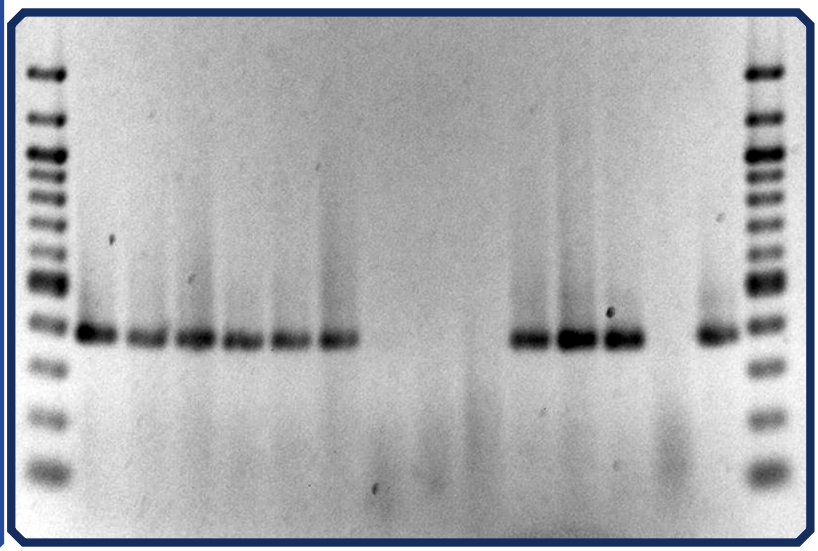
Problem

The quagga mussel (*Dreissena rostriformis bugensis*) is an introduced freshwater bivalve that is spreading across the Western United States. The mussel is negatively affecting water ecology and impacting infrastructure such as dams, water intakes, and water treatment facilities. The Reclamation Detection Laboratory for Exotic Species (RDLES) in Reclamation's Technical Service Center is dedicated to the early detection of invasive, threatened, and endangered species, and it has been responsible for advancing the science of invasive mussel early detection. Cross polarized light microscopy is the preferred and standard method for detecting mussel larvae (veligers) in raw water samples; however, degradation of the veliger shell may result in false negative microscopy findings. Veligers are microscopic, which makes identification difficult, especially if the sample contains significant amounts of sediment, organic material, or other bivalves.

RDLES has included polymerase chain reaction (PCR) testing for detecting DNA in addition to the microscopy analysis of raw water samples. This test helps reduce the likelihood of false negatives and verifies species identification, as needed. PCR is capable of detecting the presence of veligers that are degraded beyond the point of microscopic detection. PCR testing is complex and takes time to optimize because the test includes multiple steps and reagents. These complexities lead to results that can be variable and inconsistent. This Reclamation Science and Technology Program research project was designed to explain this variability.

Solution

The goal of this research study was to demonstrate how multiple factors, including DNA extraction kit type, amount of DNA in the sample, number of days to analysis, and presence or absence of DNA inhibitors, impact PCR detection of quagga mussel DNA. PCR success rate was observed in four sample scenarios, including detecting whole veliger bodies in water without inhibitors and in water with inhibitors, and detecting broken veligers, degraded veligers, and two concentrations of free-floating adult DNA in water with inhibitors.



PCR gel with positive bands indicating the presence of quagga mussel DNA in each sample.

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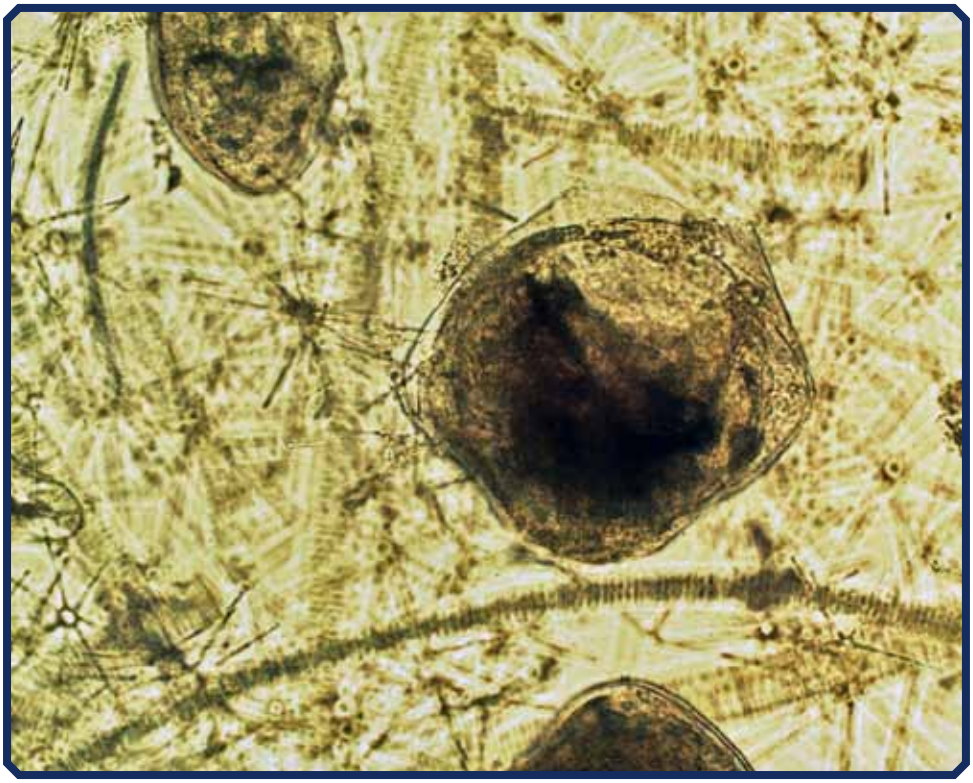
Research and Development Office
Website: www.usbr.gov/research
Telephone: 303-445-2125

Application and Results

Overall, the research study results indicate that a positive PCR result can be achieved on a water sample that previously tested negative by microscopy. This is possible because PCR can detect free-floating DNA and veligers that are degraded and broken apart. Unfortunately, many of the PCR results in this study were negative, even though a known source of DNA was present in the sample. These results suggest that early detection PCR will likely produce more false negatives than false positives.

More research is necessary to determine how water quality and chemistry affect the DNA extraction chemistries of the DNA extraction kits. While microscopy is an important aspect of *dreissenid* early detection, this research study indicates that PCR testing of *dreissenid* early detection samples is a valuable tool that is capable of detecting signs of *dreissenid* presence that would otherwise be missed by microscopy alone.



Degraded veliger not detected by cross polarized light microscopy.

Future Plans

Further research is necessary to better understand and optimize the PCR methods for detection of *dreissenid* veliger DNA in early detection samples.

"PCR testing of dreissenid mussels with early detection samples is a valuable tool that is capable of detecting signs of dreissenid presence that may otherwise be missed by microscopy alone."

Sherri Pucherelli
Biologist, Reclamation's Technical Service Center

Principal Investigators

Jamie Carmon
Former Biological Services Technician with the Technical Service Center

Jacque Keele
Biologist
303-445-2187
jkeele@usbr.gov

Sherri Pucherelli
Biologist
303-445-2015
spucherelli@usbr.gov

Denise Hosler
Botanist
303-445-2195
dhosler@usbr.gov

Environmental Applications and Research Group
Technical Service Center

Research Office Contact

Joe Kubitschek, P.E.
Invasive Mussels Research Coordinator
303-445-2148
jkubitschek@usbr.gov

More information

www.usbr.gov/research/projects/detail.cfm?id=8912

For printable version see: www.usbr.gov/research/docs/updates/2015-14-mussel-dna.pdf