# Bacterial Remediation of Microcystin HAB Toxins

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Delivering scalable freshwater HAB prevention, detection, and management technologies through collaboration, partnership, and cutting-edge science.

Problem

Cyanobacteria containing toxins have been documented in most states and are a priority concern for inland waterways. Microcystins (MCs) are the most reported freshwater toxins, with

approximately 100 analogues and are potent hepatotoxins. Conventional methods for water treatment (boiling, chlorination, and UV treatment), are unsuccessful at destroying these toxins. Naturally occurring bacteria can typically degrade MC toxins through the mlrABCD cluster. The MlrA enzyme opens the cyclic structure, and subsequently, linear toxin is  $\geq$ 160 times less toxic. MlrB further degrades the linear MC.

Objective

The goal of this project is to

reengineer and produce a shelf-stable MC degrading enzyme that can be used in the field to deactivate harmful toxins while cleanup personnel remediate MC harmful algal bloom (HAB) events in a safe environment.

### Approach

The project team will develop a

synthetically derived, bacterialproduced enzyme(s) capable of degrading MC. By producing a bicistronic MC-degrading clone, the MlrA enzyme—which linearizes the highly toxic cyclic MC, thus significantly reducing its toxicity—is coupled with the MlrB enzyme, which begins the natural degradation process by further cleaving the MC. The resultant clone expels the enzyme(s) and allows for high level production and eventual purification of the enzyme(s). The MlrAB will be purified and its effectiveness against MC in a variety of real-world matrices will be evaluated. The final product of this project is anticipated to be a shelfstable, enzyme powder that can be safely administered to MC-impacted bloom areas to reduce toxicity to wildlife and cleanup crews.

The final goal of this project is to produce a shelf-stable MC-degrading enzyme that can be used in the field to deactivate harmful toxins while cleanup personnel remediate MC-HAB events in a safe environment.

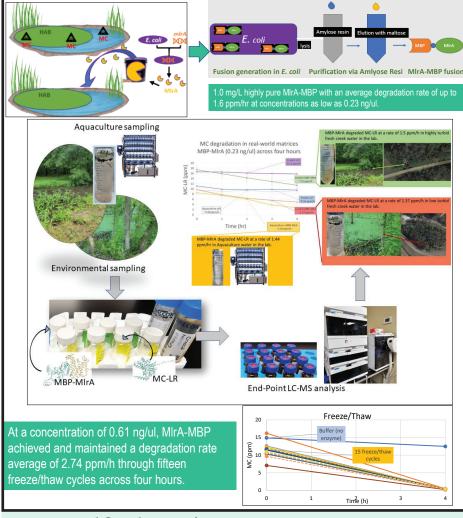


Figure 1. Workflow diagram for project.



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### Major Milestones

Date	Milestone
Publications	<b>Tech Report:</b> Jung et al. In prep. "Bacterial Remediation of Microcystin-HAB Toxins." ERDC/EL TR.
Products	<b>US Patent</b> #17405012 "Recombinant proteins having enzyme activity against microcystin and methods of water remediation" (Submitted; in review)
Tech Transfer	Industry Partner: Possible transition to Allonia, LLC
Demonstration	Meso-/field-scale demo and cost/feasibility assessment for scale-up of MlrA application (Pending)

#### Partnership/Leveraging Opportunities

This work will leverage multiple collaborations and other work units. This project involves collaboration with multiple USACE

district liaisons from Omaha, Jacksonville, and Buffalo District. In the future this project may be transferred to a partner, Allonia, LLC.

#### Value to USACE Mission

Once a bloom event occurs, detoxifying the water would be hugely beneficial to animal and human health and allow for the safety of HAB response

team personnel.



