## A Novel Biological Control Approach for Cyanobacteria

#### USACE Harmful Algal Bloom Research & Development Initiative



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

Lead: Dr. Karl J. Indest, ERDC, Karl.J.Indest@usace.army.mil

Problem Harmful algal blooms (HABs) are a worldwide problem and HABs associated cyanotoxins are a priority concern for US inland waterways. USACE district and division managers lack targeted, effective tools for combatting problematic cyanoHABs (cHABs) in their respective regions. Shortterm mitigation strategies for the effective management of HABs and their toxins are needed like target-specific novel cyanocidal chemical products.

**Objective** To identify a novel cHAB control method utilizing a bacterially derived compound(s) demonstrating both cyanocidal activity and class-level specificity that will provide USACE districts, water resource managers, and other interested stakeholders with a proven, effective alternative to less discriminatory cHAB control methods. Biological control through bacterially derived bioactive compounds remains a relatively untapped research area in cHAB mitigation, with the potential for highly specific cyanobacteria control.



**Figure 1.** Project approach to screening and indentifying natural cyanocide compounds.

Approach

In FY21, identified and evaluated experimental bacterial cyanocide compounds that achieve 30% reduction in biomass in 72 h in model organisms and in standard screening assays

on various cyanobacteria monocultures. Screened and identified aquatic/terrestrial bacteria for secondary metabolites able to inhibit algal growth in liquid/semisolid media. In FY22, there were continued algaecide evaluations on broad phytoplankton-based species and efforts to determine potential effects on nontarget species via zooplankton toxicity studies. Research efforts included partial purification and characterization of bacterial algaecide compounds. In FY23, microcosm community impact evaluations of bacterial algaecide compounds were performed.



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**Figure 2:** Flow chart of the methods used to identify new cyanocide compounds: (a) bacteria strains were tested on Microcystis sp. and those that reduced the algal biomass by 30% were used for downstream experiments; (b) candidate bacterial strains were extracted and extracts analyzed using flash chromatography; (c) purified standards were run on a Nuclear Magnetic Resonance (NMR) instrument to help identify unknown compounds; (d) bacterial extracts were screened for cyanocide activity represented as zones of clearing on a cyanobacterial lawn plate.

## **Major Milestones**

Deliverable	Description
Publications	<ul> <li>Journal Article: Lefler et al. 2022. "Draft Genome Sequences of Bacteria with Cyanocidal Potential Isolated from Eutrophic Waters Associated with Cyanobacterial Harmful Algal Blooms." <i>Microbiology Resource Announcements</i>. https://doi.org/10.1128/mra.00205-23.</li> <li>Journal Article: (In Prep) Lefler et al. "Efficacy and Toxicity of Bacterial Derived Cyanocidal Compounds on Target and Nontarget Species."</li> </ul>

## Partnership/Leveraging Opportunities

This work will leverage a collaboration with the University of Florida's Fort Lauderdale Research and Education Center

(FLREC) via Dr. Dail Laughinghouse. Currently, this project supports a part-time ORISE scientist under the direction of Dr. Laughinghouse. In addition, FLREC is a unique laboratory resource that cultures and maintains several environmentally relevant HABs and other algal species needed for successful execution of this work unit.

## Value to USACE Mission

USACE District and Division managers lack targeted, effective tools in combatting problematic localized cHABs in their respective regions. Target-

specific novel cyanocidal chemical products offer short-term mitigation strategies for effective management of HABs and their toxins.



