

Determining how the ecophysiology of different *Microcystis* strains underpins succession and toxicity of harmful cyanobacterial blooms in Lake Erie

1. Title Page Information

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- e) Abstract:

Blooms of the toxic cyanobacterium *Microcystis aeruginosa* threaten ecosystems and freshwater drinking supplies in the Great Lakes and around the world. This project will fill a major knowledge gap that impedes our ability to deal with these blooms by determining environmental and microbiological controls on bloom toxicity. We will characterize the physiology and spatiotemporal distribution of *Microcystis* strains in Lake Erie and integrate this knowledge into an ecological model to test the hypothesis that observed dynamics of bloom toxicity are due to seasonal ecological succession of strains that are differentially adapted to temperature, light, nitrogen availability, and oxidative stress. This research will advance the field towards the ability to model, predict, and forecast the toxicity of harmful cyanobacterial blooms and to develop science-driven policies to prevent them. Thus, this project directly addresses Michigan Sea Grant focuses areas of *Resilient Communities and Economies* and *Healthy Coastal Ecosystems*. The research will train a graduate student and four undergraduate students, including those from underrepresented backgrounds, and be integrated into courses reaching hundreds of undergraduate students per year, thus contributing to the *Environmental Literacy and Workforce Development* focus area of the Michigan Sea Grant Strategic Plan.

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PROJECT NARRATIVE

3. Introduction

Statement of Research Problem

Blooms of the toxic cyanobacterium *Microcystis aeruginosa* threaten ecosystems and drinking water in Lake Erie, Lake St. Clair, Green Bay, Saginaw Bay, and temperate lakes globally. These blooms have been increasing in frequency and intensity for decades and are projected to strengthen further in the face of climate change and increased intensity of agriculture. Though the massive impact on Great Lakes ecology has been long recognized, the gravity of this issue for human health and drinking water security was highlighted by the 2014 crisis in Toledo, Ohio, during which drinking water was shut down for three days due to the conspiracy of physical and biological events during a toxic bloom (Steffen et al., 2017).

Phosphorous from agricultural runoff has been identified as the main cause of bloom *biomass*, but the drivers of bloom *toxicity* are still debated. Toxicity, in terms of quantity of the main toxin (microcystin) produced per bloom biomass, varies widely depending on the time and place of the bloom. A growing body of evidence indicates that environmental factors such as nitrogen availability, temperature, and resistance to oxidative stress determine this variability in toxicity (Otten and Paerl, 2011; Paerl and Otten, 2013; Cory et al., 2016; Gobler et al., 2016; Paerl et al., 2016; Peng et al., 2018; Newell et al., 2019). However, the mechanisms that explain the biological and environmental drivers of bloom toxicity are have not been identified. Understanding these mechanisms is required for the development of predictive models that can (i) provide early warning systems for coastal communities at risk and (ii) determine effective policies for the regulation of nutrient loading in order to protect Great Lakes ecosystems and natural resources.

In Lake Erie and temperate lakes around the world, temporal patterns of *Microcystis* blooms suggest that the composition of *Microcystis* strains within the blooms is a major determinant of bloom toxicity (Kurmayer et al., 2003; Davis et al., 2009; Gobler et al., 2016; Jankowiak et al., 2019; Smith et al., 2019). The early phase of the bloom is dominated by toxic *Microcystis* strains that carry the *mcy* genes for biosynthesis of the toxin (microcystin), whereas later phases are dominated by strains that lack these genes (**Fig. 1**).

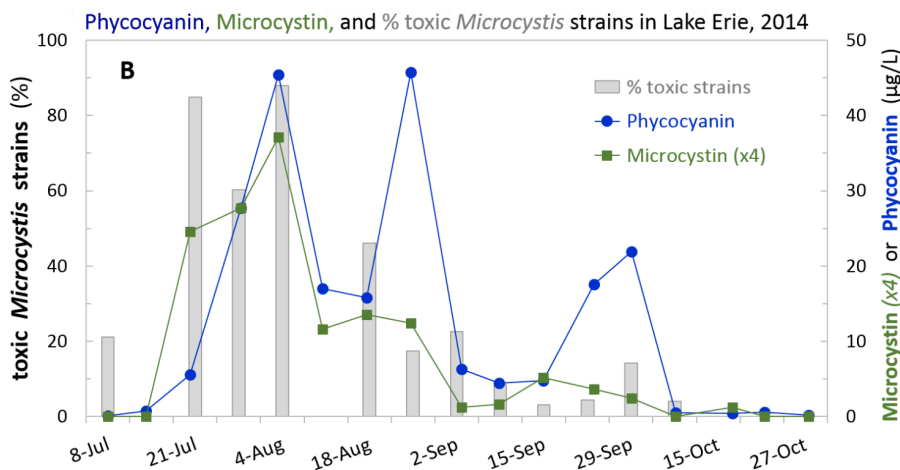


Fig. 1. Concentrations of phycocyanin (a cyanobacterial pigment, proxy for biomass) and microcystin (toxin), and the ratio of toxic to non-toxic *Microcystis* strains (as a %) in Lake Erie in 2014. From Smith et al. (2019).

Field and laboratory data suggest that toxic strains predominate at high light, nitrate availability, and oxidative stress, whereas non-toxic strains predominate at low light and oxidative stress and when organic forms of nitrogen predominate (Kardinaal et al., 2007; Davis et al., 2009; Davis et al., 2010; Dziallas and Grossart, 2011; Zilliges et al., 2011). However, despite their critical impacts on human and ecosystem health, little is known about the biology of these different strains of *Microcystis*. Field studies have limited resolution in determining the phenotypic properties of strains. Lab studies, on the other hand, are typically disconnected from the real natural systems, thus it is unclear if insights from the lab are applicable to bloom dynamics in the field.

One of the major causes for concern about the link between field and lab studies of *Microcystis* is the biological diversity of its different strains and species. Recent genetic studies of the genus show that there are 4-8 distinct phylogenetic clades, some of which correspond to species defined by cell morphology. However, *Microcystis aeruginosa* is paraphyletic, with different strains falling in distinct clades (Perez-Carrascal et al., 2019). Furthermore, these different clades harbor distinct genes that are present and expressed in the environment (Meyer et al., 2017), some of which appear to be locally transferred among strains (Perez-Carrascal et al., 2019). Together these findings suggest that there are locally adapted strains and/or species of *Microcystis*, raising the possibility that biological properties of *Microcystis* from one location may not apply to *Microcystis* at another location. This observed genetic and biogeographical diversity of *Microcystis* has not yet been linked to ecophysiological diversity of *Microcystis*, i.e., adaptations of strains to environmental and ecological factors such as light, concentration and form of nutrients, deterrence of grazers, and resistance to viruses.

Much of our knowledge about the physiological diversity of *Microcystis aeruginosa* comes from studies that compare toxic and non-toxic strains. Non-toxic strains are able to out-compete toxic strains at low light (Kardinaal et al., 2007). Toxic strains out-compete non-toxic strains for CO₂ (Van de Waal et al., 2011) and at higher oxidative stress (Dziallas and Grossart, 2011; Zilliges et al., 2011). These findings have been used to suggest tradeoffs in production of toxin versus other physiological traits. However, toxicity (i.e., presence of *mcy* genes and/or ability to produce microcystin) is paraphyletic trait (Berry et al., 2017; Perez-Carrascal et al., 2019), and examination of more strains often shows that ability to produce toxin does consistently correlate with phenotypes such as carbon uptake (Sandrini et al., 2014). Moreover, there are differences in phenotypes such as light-dependent growth among toxic strains (Bottcher et al., 2001; Hesse and Kohl, 2001). Likewise, studies of *Microcystis* in our laboratory show that phenotypes vary among different strains of toxic and non-toxic *Microcystis* (**Fig. 2**). Finally, the proteome of *Microcystis* (i.e., total content of all proteins within a cell/culture) varies widely between strains and does not reflect toxicity (Alexova et al., 2011). Overall, these results indicate that the ability to produce the toxin is not a main determinant of strain-level ecophysiological traits (i.e., growth response to environmental factors) nor an appropriate means of classifying strains. Thus, the strain-level ecophysiological drivers of bloom dynamics remain poorly understood, and in no case have strain dynamics in a real bloom been directly linked to physiological knowledge of those strains derived from pure culture. In particular, despite their extraordinary impact on human and environmental health, little is known about the biology of Great Lakes strains of *Microcystis* (Meyer et al., 2017). We assert that **this lack of knowledge about the diversity and biology of *Microcystis* strains, especially Great Lakes strains, is a major impediment to understanding and predicting bloom toxicity.**

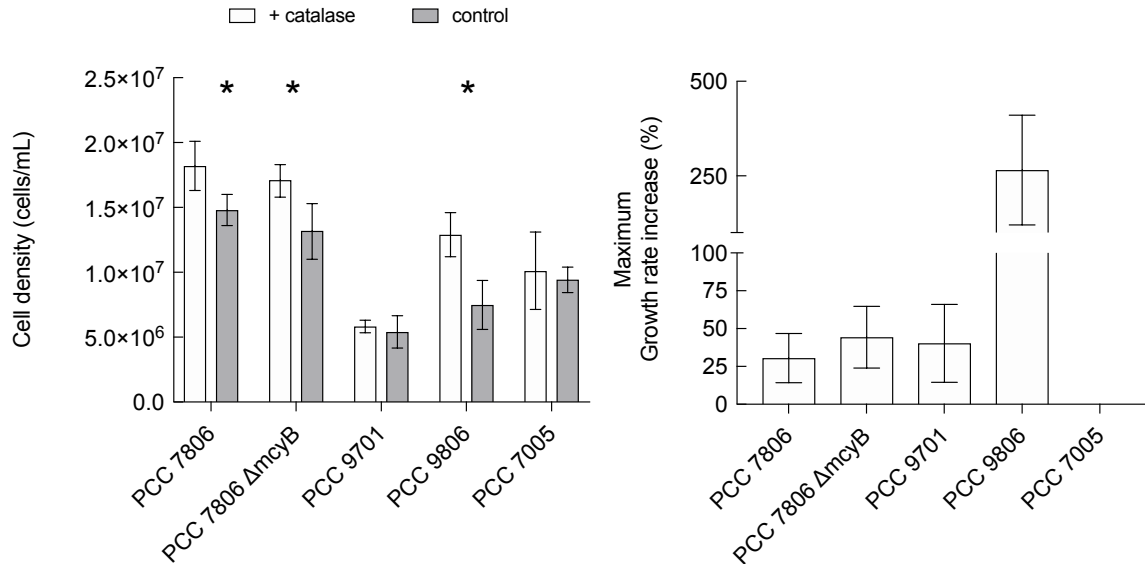


Fig. 2. Maximum cell densities (left) and growth rate increase in the presence of catalase (right), an enzyme that decomposes hydrogen peroxide and alleviates oxidative stress, in lab cultures of different strains of *Microcystis aeruginosa*. The asterisks on the left indicate cultures that had a significant difference between the treatment and control groups as determined by a one-tailed Welch's T-test. Error bars show 95% confidence intervals. The results show that different strains of *Microcystis* have different sensitivity/resistance to oxidative stress, but not in the simple toxic vs. non-toxic manner hypothesized previously (Paerl and Otten, 2013).

Here we propose a project that will link cultured Lake Erie *Microcystis* strains to those in the field, reveal their biological traits, and integrate this knowledge into ecological models of the Western Basin of Lake Erie to test whether these traits are sufficient to explain observed bloom dynamics. While there are many traits that influence the success of *Microcystis* strains, here we propose to focus on the responses to light, oxidative stress, temperature, and nitrogen availability. Our focus on these conditions reflects that they are increasingly linked to bloom dynamics (Paerl and Otten, 2013; Cory et al., 2016; Gobler et al., 2016; Newell et al., 2019), show evidence of interactions (i.e., response to light depends on nitrogen concentration) (Chaffin et al., 2018), and can be feasibly studied within the temporal and financial constraints of this project. The proposed research will build on, integrate with, and leverage current modeling efforts from a NOAA CIGLR Postdoctoral Fellowship (see letter of support from Dr. Bradley Cardinale, Director of CIGLR), a NOAA Ecohab Project (see letter of support from Dr. Justin Chaffin, PI, Stone Laboratory), and genetic data produced by grants from the National Science Foundation and National Institutes for Environment, Health, and Safety (see letter of support from Dr. George Bullerjahn, Director of the NIEHS/NSF Great Lakes Center for Fresh Waters and Human Health). Overall, this project will move us towards the capability of accurately forecasting toxicity of harmful cyanobacterial blooms in the Great Lakes.

Research Relevance: How this research addresses current issues and Michigan Sea Grant Strategic Plan

This project will produce critical information about the diversity and biology of *Microcystis aeruginosa*, the dominant cyanobacterium responsible for toxic blooms that threaten ecosystems and drinking water supplies around the Great Lakes. This research advance is highly relevant to

at least two focus areas of the Michigan Sea Grant Strategic Plan. First, because the project will enable the development of models that are able to predict the toxicity of harmful algal blooms (a capability that does not currently exist), this project is relevant to the *Resilient Communities and Economies* focus area (particularly outcomes 6.2, 6.8, 8.1, 8.6, 8.7, 9.1, and 9.5). Predictive models will enable forecasts that can warn drinking water managers in advance and provide guidance to charter boat captains (a \$1 billion industry) regarding areas to avoid. Second, the project will enable detailed studies of the relationship between nutrient loading (especially nitrogen, a critical yet heretofore unregulated form of fertilizer) and toxic blooms, thus guiding science-driven policies and contributing to the *Healthy Coastal Ecosystems* focus area (particularly outcomes 1.1, 1.2, 1.3, 2.1, 2.4, 2.8, and 3.3). “Supporting research projects that develop information to guide decisions about complicated environmental problems” and “water quality/contamination” were among the highest priority issues identified by respondents to the Michigan Sea Grant Stakeholder Survey. Improvements in modeling would feed into the MISG performance measures of “Sea Grant tools and technologies used by partners/customers to improve ecosystem-based management” and “peer-reviewed publications”.

In addition to the relevance of the scientific outputs as described above, the research will also contribute to the *Environmental Literacy and Workforce Development* focus area of the Michigan Sea Grant Strategic Plan. First, the project will train one graduate student and four undergraduate students in methods of limnology and environmental microbiology; these skills are in high demand for water quality professionals (11.1, 11.2, 11.3, 11.4, 11.5, 11.6). Second, results, pictures, and videos from the project will be integrated into a large format course taught by the PI at the University of Michigan (outcomes 10.4, 10.9). This course (EARTH 112, *The Great Lakes*) has enrollment of 200-400 undergraduates *per semester* and is a wonderful opportunity to develop environmental literacy among Michigan residents.

Finally, this project supports core values and cross-cutting principles defined in the MI Sea Grant Strategic Plan. Advancement of next-generation models for forecasting bloom toxicity represents an innovative solution to a serious threat to human and ecosystem health. This project would support a collaborative partnership between academia, the federal government, and the private sector. It would engage high school, undergraduate, and graduate students in Great Lakes sustainability issues and enhance diversity and inclusion by integrating students from underrepresented backgrounds.

4. Project Description

Response to Reviews of the Pre-proposal

We appreciate reviewer feedback on the pre-proposal, which was positive overall, and have carefully considered it in revising and expanding this full proposal. One reviewer requested more information about the culturing methods (*Objective 1*), and another expressed a concern that comparing the model to observations and refining the model accordingly may be too ambitious (*Objective 5*). We have now removed *Objective 1* due to a crucial update: we now have a sufficient number of cultured *Microcystis* strains such that new isolations are no longer necessary; this reduces the ambition of the project and improves the feasibility of the proposed timeline. *Objective 4* has been updated to provide more details and preliminary results and to remove the model refinement portion of this goal. However, the modeling and comparison to

field observations is retained as we feel that this provides a critical link between the field and laboratory data.

Project Goals and Objectives

The overall goal of this project is to determine the physiological traits of various *Microcystis* strains that govern their observed seasonal succession during Lake Erie blooms, thus underpinning bloom toxicity. Our **hypothesis** is as follows:

The seasonal succession of toxic to non-toxic Microcystis strains is due to distinct adaptations to four intertwined environmental factors: temperature, availability of various forms of nitrogen, light intensity, and resistance to oxidative stress (i.e., environmental H₂O₂).

Thus, these differential adaptations would define distinct niches for different strains, reflecting tradeoffs between traits. To assess this hypothesis we will test the predictions that (i) early bloom strains are characterized by relatively faster growth at high temperature, light, and oxidative stress, with inorganic forms of nitrogen (*i.e.*, nitrate), and (ii) late bloom strains grow better at lower temperature and light, with organic forms of nitrogen (*e.g.*, urea), and are more sensitive to oxidative stress. The **specific objectives** of the project are as follows:

- 1) Identify which cultured strains are representative of dominant bloom strains.
- 2) Characterize the growth of representative strains with respect to temperature, light, oxidative stress, and availability/concentration of nitrogen species.
- 3) Incorporate the newly defined growth parameters into a coupled hydrodynamic-ecological model to simulate bloom strain composition in space and time, and compare model results to field observations.
- 4) Communicate and disseminate results of the project.

Research Methods

Our objectives will be achieved with the following methods:

Objective 1: Identify which cultured Lake Erie strains are representative of dominant blooms strains.

This effort will build on our culturing efforts from 2017 and 2018, which produced 43 newly cultured strains from Lake Erie. *Microcystis* strains were cultured NOAA GLERL's weekly monitoring cruises, which we have participated in every week since 2014. This included early, middle, and late stages of the bloom (**Fig. 1**). Individual *Microcystis* colonies were picked, washed, and inoculated into sterile media (BG-11 and WC medium, both full strength and adjusted to nutrient concentrations that are closer to environmental relevance) in liquid and on solid agar plates. Several strains are already axenic (no other bacteria present), and the rest are still undergoing successive transfer plating and/or dilution to extinction, and filtered with 3 µm filters (to remove smaller bacteria) to obtain axenic cultures.

To genetically identify these cultures and assess which are most abundant in the populations of Western Lake Erie, DNA from (i) individual cultured strains and (ii) weekly field samples from 2014-2019 will be extracted and the 16S-23S rRNA internal transcribed spacer (ITS) will be amplified by polymerase chain reaction and sequenced as described previously (Otten et al., 2017). This gene is conserved across all *Microcystis* species yet provides sufficient resolution for

distinguishing closely related strains (Otten et al., 2017)(Janse_04). The abundance of cultured strains in Lake Erie blooms will then be quantified by two methods: (i) BLAST search (Altschul et al., 1990) using the ITS sequences from cultures as a custom database and the sequences retrieved from field samples as the query, and (ii) oligotyping of ITS sequences as we have done for 16S rRNA gene sequences (Berry et al., 2017). The five strains that show greatest abundance will be further characterized in experiments described below. Previous results based on a fragment of the 16S rRNA gene showed three dominant types of *Microcystis* in the 2014 bloom (Berry et al., 2017), but we expect the 16S-23S rRNA sequence to provide a more highly resolved picture of strain variation.

Objective 2: Characterize the growth of representative strains with respect to temperature, light, oxidative stress resistance, and availability of nitrogen species.

The growth rates of five cultured strains that are most abundant in Lake Erie blooms (as determined by *Obj. 1*) will be characterized in modified BG-11 medium (Stanier et al., 1971) in a multi-factorial experimental design (which is critical for microbial ecology (Duarte et al., 1997) especially for the interacting factors studied here; see below) under three different temperatures, light intensities, and levels of oxidative stress, and nine different nitrogen regimes, as follows:

Table 1. Experimental conditions for growth of *Microcystis* strains.

Temp.			Light			Nitrogen source									Oxidative stress*		
°C			umol photons m ⁻² /s ⁻¹			NO ₃ (μM)			NH ₄ ⁺ (μM)			Urea (μM)			H ₂ O ₂		
21	24	27	200	400	800	5	20	100	1	5	10	0.1	2	10	0	amb	high

*Oxidative stress is induced with hydrogen peroxide (H₂O₂), which occurs naturally in Lake Erie (Cory et al., 2016) and inhibits *Microcystis* growth (Zilliges et al., 2011). Conditions are established as follows: “0”, amended with catalase, which decomposes H₂O₂ to non-detectable concentrations; “amb”, ambient culture conditions, where H₂O₂ concentrations are 600-1200 nM, in the range where *Microcystis* is slightly inhibited; “high” is achieved by adding H₂O₂ three times per days, aiming to maintain concentrations at 2 μM, near the maximum measured in early/peak blooms of Lake Erie (Zilliges et al., 2011).

These experimental conditions were chosen based on current knowledge of factors that favor toxic versus non-toxic strains, and to represent the range of conditions experienced by Lake Erie blooms. Manipulation of hydrogen peroxide (H₂O₂) concentrations will *test the hypothesis that oxidative stress, especially environmental H₂O₂, favors toxic strains of Microcystis* (Dziallas and Grossart, 2011; Paerl and Otten, 2013) due to the protective effect of microcystin (Zilliges et al., 2011). There is a peak of H₂O₂ concentration at the onset of Lake Erie blooms annually, coincident with peak toxicity and peak levels of toxic strains (**Fig. 1**), consistent with oxidative stress selecting for toxic strains. We will vary temperature because it has been shown to modulate sensitivity of cyanobacteria to H₂O₂ (Ma et al., 2017) and different strains have different temperature adaptations (Bui et al., 2018).

Manipulation of nitrogen will test the *hypothesis that different strains of Microcystis have differential adaptations to the concentration and form of nitrogen*. While this has been suggested based on evidence from field observations and mecosm experiments (Davis et al., 2010),

including those in Western Lake Erie (Gobler et al., 2016) it has not been directly tested on cultured strains from Lake Erie. Light also affects competition between *Microcystis* strains (Kardinaal et al., 2007) and interacts with oxidative stress and nitrogen availability (Chaffin et al., 2018).

Each condition will be run in triplicate in 10-ml volumes in glass test tubes, for a total of 3,645 test tubes for the five strains. We will run one light/temperature regime at a time (i.e., nine separate experiments, 405 tubes per experiment) to calculate growth rates over the course of ~10 day growth curves in incubators with controlled light and temperature. At two weeks per experiment, these growth experiments will take approximately 18 weeks in total. Growth will be tracked by cell counts with a hemacytometer. Instantaneous growth rates at each day of the experiment will be calculated by fitting the cell counts to a Gompertz growth model, and taking the derivative of the growth function (Zwietering et al., 1990).

Objective 3: Incorporate the newly defined growth parameters into a coupled hydrodynamic-ecological model to simulate bloom strain composition in space and time, and compare model results to field observations.

A critical step in linking our field and laboratory data and testing our hypotheses will be to integrate the newly defined growth parameters from the lab into a coupled hydrodynamic-ecological model to simulate bloom dynamics, make predictions about the spatiotemporal distribution of specific strains, and compare these model results to observations from the field. For this task we will use the WLEEM model, an integrated hydrodynamic/ecological model of the Western Basin of Lake Erie (Verhamme et al., 2016). This model explicitly represents five phytoplankton groups and their key traits including growth rate as a function of temperature, nutrient concentration and ratio, and light intensity. Originally the phytoplankton functional groups in this model included cyanobacteria, diatoms, and green algae; we recently modified it to include multiple different classes (i.e., strains) of *Microcystis*. Growth characteristics for each strain, as defined in *Obj. 2*, will be used parameterize two classes of *Microcystis* (representing strains) for variables relating to growth temperature, half saturation constant for nitrogen, and saturating light intensity (see Verhamme et al. 2016). Oxidative stress will be implemented as a H_2O_2 concentration-dependent growth rate penalty. Model outputs are in terms of biomass of the different strains, and our preliminary work has shown that subtle differences in parameters such as temperature optimum can have substantial impacts on the strain composition of blooms (Fig. 3).

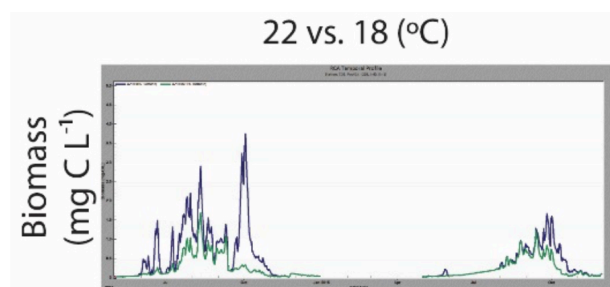


Figure 3. WLEEM model output for two phytoplankton classes where all physiological parameters were identical except for Optimum Growth Temperature (TOPT). Models were run for a 2-year period (2014-2015). Green lines represent the control group where TOPT was set at 22°C, Blue lines indicate experimental group where TOPT was set at 18°C. This shows that subtle differences in strain physiology can have large impacts on the *Microcystis* strain composition of blooms, but the physiological traits of Lake Erie *Microcystis* strains remain unknown. The horizontal axis is time, with biomass occurring between July and October of 2014 and 2015.

We will run the model with two *Microcystis* strains at a time (given the limited number of phytoplankton classes available). Model runs simulating full bloom seasons (May – November) will be run for years in which we produced genetic data in *Obj. 2* (2014-2018), using observed environmental conditions and nutrient loadings from those years. Model outputs will be analyzed in two ways: (i) at specific locations (monitoring stations where samples were collected) for direct comparisons to genetic data from those locations and (ii) in a basin-integrated manner to remove effects due to spatial variability, for which model resolution may be limited. Model results will be directly compared to genetic data from field samples in terms of (i) biomass of *Microcystis* and (ii) percentage of strains carrying *mcy* genes for microcystin biosynthesis. Sensitivity analyses will be conducted to determine which parameters are most influential in model output, and Bayesian inference will be used to evaluate the most probable scenarios to explain field observations. This model has limitations (e.g., only five phytoplankton classes; does not explicitly represent all forms of nitrogen), but it will allow us to quantitatively test whether the laboratory-measured traits can feasibly be responsible for observed shifts in the strain composition of Lake Erie *Microcystis* blooms. Further, results will help guide development of next generation models, which will include more phytoplankton classes and more realistic modeling of nutrient dynamics.

Objective 4: Communicate and disseminate results of the project.

The results of this project will be disseminated widely. We will present results at the International Association of Great Lakes Research annual conference on Great Lakes Research and at regional meetings, e.g., the Michigan Geophysical Union, CIGLR partner's meeting, Undergraduate Research Opportunities Program symposium, and Water@Michigan meeting, which has a wide-ranging audience including water managers and practitioners. Results will also be published in at least one peer-reviewed journal. Raw data, including the newly defined growth parameters, linked to gene sequences, will be made publicly available via a database of *Microcystis* traits. In the long run this is part of our broader effort, with pending support from NOAA and in collaboration with CIGLR and GLERL, to synthesize data on *Microcystis* physiological parameters and to integrate it with genetic data in a "Microcystis pheno-genotype database". As a part of the current project, we will make all physiological data publicly available through a website (hosted at <https://sites.lsa.umich.edu/geomicro/databases/>). This database will be searchable by strain name or parameter (i.e., return all strains for which we have growth rate data on nitrate concentration) and will also include download options for spreadsheets of all data.

Applicability and users of the information to be developed

The direct and immediate users of the data will be scientists and engineers who are studying harmful cyanobacterial blooms in Lake Erie and/or developing next generation models for predicting their toxicity. This potentially includes academic scientists, scientists and water quality professionals at federal agencies (NOAA, EPA, USGS), state agencies (e.g., Michigan Department of Environmental Quality), and companies (e.g., Limnotech). The results of the project will also be used more widely by such entities around the nation and world as ecological models of *Microcystis* blooms are advanced at multiple levels (Dick, 2017), from trait-based ecological modeling (Litchman and Klausmeier, 2008) to mechanistic molecular models (Hellweger et al., 2016), to next-generation water quality models that are widely used by water quality professionals.

5. Project Timeline

The timeline for this project is shown below in **Fig. 4**. We are prepared to start the project Feb 1, 2020, and to complete the proposed research in two years.

Obj.	Task	Leader	Support	Year 1				Year 2			
				Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4
1	Extract DNA	Yancey	Dick	■							
1	PCR of 16S-23S rRNA	Yancey	Dick		■						
1	Sequence PCR products	Yancey	Dick		■						
2	Growth Experiments	Yancey	UGs	■	■	■	■				
3	Modeling runs	Yancey	Verhamme		■	■	■	■	■	■	
3	Model sensitivity analysis	Yancey	Stow		■	■	■	■	■	■	
3	Compare model & field results	Yancey	Dick			■	■	■	■	■	
4	Build <i>Microcystis</i> database	Yancey	Dick, UGs	■	■	■	■	■	■	■	
4	Regional presentations	Yancey	all		■	■	■	■	■	■	
4	IAGLR presentation	Yancey	all					■	■	■	
4	Peer-review publication	Yancey	all								■

Figure 4. Timeline of research tasks by project quarter. Project tasks, team leader, and support are identified for each task. “UGs”, undergraduate students.

6. Overview of the Research Team

Relevance of PI's previous accomplishments to this project

Dr. Gregory Dick is an Associate Professor in the Department of Earth and Environmental Sciences, with promotion to Professor anticipated in 2020. He is an experienced leader of scientific projects, having previously led 11 funded grants as Principal Investigator, including several large, collaborative, multi-investigator projects. He has published 62 peer-reviewed articles in top scientific journal including *Science*, *Proceedings of the National Academy of Sciences*, *Nature Communications*, and *Nature Reviews Microbiology*. These papers have been cited well over 4,000 times. These accomplishments have earned recognition as an Alfred P. Sloan Foundation Fellow in Ocean Sciences.

Prof. Dick's expertise is in microbial ecology, environmental “omics”, and biogeochemistry. Over the past five years his research focus has shifted from the geomicrobiology of deep-sea hydrothermal vents towards harmful cyanobacterial blooms. He has extensive experience in microbial physiology, environmental molecular microbiological methods, and integrating microbial information into ecological models, thus he is well qualified to lead the proposed project. Prof. Dick is also accomplished as a teacher, winning the 2015 U-M LSA Individual Award for Outstanding Contributions to Undergraduate Education and the 2019 Geoclub “Best Professor” awards. He has engaged numerous regional and national media outlets, including *National Public Radio*, to broadly disseminate the results of his research. He is also engaged in diversity, inclusion, and equity issues as a PI of an NSF-funded project on “Enhancing pathways into the Geosciences” and the faculty advisor for Earth Camp, a summer experience for high school students from underrepresented backgrounds. Thus, he is well prepared to work towards the outreach and diversity, equity, and inclusion goals of this project.

Roles and responsibilities of team members

Dr. Gregory J. Dick will lead the entire project, supervise PhD student Colleen Yancey and the undergraduate students, and participate in the analysis, interpretation, writeup, and presentation of results. He will hold weekly meetings with the PhD student and undergraduate student and coordinate monthly meetings with the entire team.

Colleen Yancey, currently a first-year PhD student in the Department of Earth and Environmental Sciences at the University of Michigan, will be the primary researcher who conducts the laboratory experiments, DNA sequencing, and modeling. This project will be the focus of her second and third years of the PhD program and it will be one of the main thrusts of her dissertation research. Yancey is proficient at culturing *Microcystis* and in laboratory and computational methods of molecular microbiology approaches. She will also work with the PI and the rest of the team to prepare manuscripts and presentations.

Ed Verhamme is a Project Engineer with LimnoTech who has been involved in dozens of real-time observations and applications of hydrodynamic, water quality, and nutrient models, including the WLEEM model, to study the Great Lakes. Verhamme will assist Yancey in running the WLEEM model.

John Bratton is a Senior Scientist at LimnoTech with broad expertise in earth and environmental sciences. He has extensive expertise in Great Lakes harmful cyanobacterial blooms, and the WLEEM model and will advise the team on overall research and modeling strategies.

Craig Stow is a Research Scientist at the NOAA Great Lakes Environmental Research Lab. He has over 25 years of experience in using ecological modeling, uncertainty analysis, and Bayesian inference to studying biogeochemical cycling and ecology in aquatic ecosystems. Dr. Stow will advise the PhD student (Yancey) in the analysis of model output and field data via sensitivity analyses and Bayesian inference.

Undergraduate Students are deeply involved in research in PI Dick's lab and often produce substantive results that earn them authorship on peer-reviewed scientific publications, even as first-author (e.g., (Den Uyl et al., 2016; Marcus et al., 2017; Sharrar et al., 2017)). The lab currently hosts five undergraduate students that have been trained in the methods that will be employed for this project, including making media, culturing cyanobacteria, conducting laboratory experiments, counting cells, and determining growth rates. This includes one African American student (Ariana Mann) and three students (Claire Zwiers, Will James, and Raina Ruman) that are rising sophomores and are expected to stay in the lab and complete research-based undergraduate theses based on the current project. Additional students will be recruited from the Undergraduate Research Opportunities Program and from Prof. Dick's Earth Camp program as needed, with particular effort in recruiting students from underrepresented backgrounds. These students are expected to conduct lab experiments, analyze data, and participate in the write-up of publications and presentation of results.

Previous experience of this team working together

Dick, Stow, Bratton, and Verhamme previously worked together in advising Dr. Kevin Meyer, who was the *CIGLR Postdoctoral Fellow* from 2018-2019. The current proposal is a direct outgrowth of that project, and it would continue to support this collaboration between academic, federal, and corporate scientists.

7. References

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- Chaffin, J.D., Davis, T.W., Smith, D.J., Baer, M.M., and Dick, G.J. (2018) Interactions between nitrogen form, loading rate, and light intensity on *microcystis* and *planktothrix* growth and microcystin production. *Harmful Algae* **73**: 84-97.
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- Ma, L., Calfee, B.C., Morris, J.J., Johnson, Z.I., and Zinser, E.R. (2017) Degradation of hydrogen peroxide at the ocean's surface: The influence of the microbial community on the realized thermal niche of prochlorococcus. *ISME J*.
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- Meyer, K.A., Davis, T.W., Watson, S.B., Deneff, V.J., Berry, M.A., and Dick, G.J. (2017) Genome sequences of lower great lakes microcystis sp. Reveal strain-specific genes that are present and expressed in western lake erie blooms. *PLoS One* **12**: e0183859.
- Newell, S.E., Davis, T.W., Johengen, T.H., Gossiaux, D., Burtner, A., Palladino, D., and McCarthy, M.J. (2019) Reduced forms of nitrogen are a driver of non-nitrogen-fixing harmful cyanobacterial blooms and toxicity in lake erie. *Harmful Algae* **81**: 86-93.
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- Peng, G., Martin, R.M., Dearth, S.P., Sun, X., Boyer, G.L., Campagna, S.R., Lin, S., and Wilhelm, S.W. (2018) Seasonally relevant cool temperatures interact with n chemistry to increase microcystins produced in lab cultures of microcystis aeruginosa nies-843. *Environ Sci Technol* **52**: 4127-4136.
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8. Qualifications

CURRICULUM VITAE: Gregory J. Dick

A. PROFESSIONAL PREPARATION

University of Virginia, Biology	B.A., 2000
Scripps Institution of Oceanography, UCSD, Marine Biology	Ph.D., 2006
University of California, Berkeley, Metagenomics	Postdoc, 2007-2008

B. APPOINTMENTS

Associate Chair for Curriculum and Undergraduate Studies, Dept. of Earth & Environmental Sciences, U. Michigan	2016 – present
Associate Professor – Dept. of Earth & Environmental Sciences and Dept. of Ecology & Evolutionary Biology, U. Michigan	2014 – present
Assistant Professor – Dept. of Earth & Environmental Sciences and Dept. of Ecology & Evolutionary Biology, U. Michigan	2011 – 2014

C. RECENT HONORS AND AWARDS

Best Professor, Dept of Earth & Environmental Sciences Geoclub	2019
Individual Award for Outstanding Contributions to Undergraduate Education, College of Literature, Science, and Arts, University of Michigan	2015
Alfred P. Sloan Research Fellow in Ocean Sciences	2013

D. PUBLICATIONS (<https://scholar.google.com/citations?user=76nnaMsAAAAJ&hl=en>)

* = student advisee; ** = postdoc advisees; underline = Dick lab member

Most closely related to the proposed project

Kharbush J, DJ Smith*, M Powers, HA Vanderploeg, D Fanslow, RL Robinson, **GJ Dick**, and A Pearson (2019). Chlorophyll nitrogen isotope values track shifts between cyanobacteria and eukaryotic algae in a natural phytoplankton community in Lake Erie. *Organic Geochemistry* 128: 71-77.

Chaffin JD, TW Davis, DJ Smith*, MM Baer, and **GJ Dick** (2018). Interactions between nitrogen form, loading rate, and light intensity on *Microcystis* and *Planktothrix* growth and microcystin production. *Harmful Algae*, 73: 84-97.

Driscoll CB, KA Meyer**, S Šulčius, NM Brown, **GJ Dick**, H Cao, G Gasiūnas, A Timinskas, Y Yin, ZC Landry, TG Otten, TW Davis, SB Watson, and TW Dreher (2018). A closely-related clade of globally distributed bloom-forming cyanobacteria within the Nostocales. *Harmful Algae* 77: 93-107.

Kramer BJ, TW Davis, KA Meyer**, BH Rosen, JA Goleski, **GJ Dick**, G Oh, and CJ Gobler (2018). Nitrogen limitation, toxin synthesis potential, and toxicity of cyanobacterial populations in Lake Okeechobee and the St. Luci River Estuary, Florida, during the 2016 State of Emergency event. *PLoS One* 13: e0196278.

Dick GJ (2018). Genomic Approaches in Earth and Environmental Sciences. Wiley-Blackwell, 12 chapters, 176 pages. ISBN: 978-1-118-70824-8

- Dick GJ** (2017). Embracing the mantra of modelers and synthesizing omics, experiments, and models. *Environ. Microbiol.*, 9: 18-20.
- Meyer KA**, TW Davis, SB Watson, VJ Denef, MA Berry, and **GJ Dick** (2017). Genome sequences of lower Great Lakes *Microcystis* sp. reveal strain-specific genes that are present and expressed in western Lake Erie blooms. *PLoS One*, 12: e0183859.
- Steffen MM, TW Davis, RM McKay, GS Bullerjahn, LE Krausfeldt, JMA Stough, ML Neitzey, NE Gilbert, GL Boyer, TH Johengen, DC Gossiaux, AM Burtner, D Palladino, M Rowe, **GJ Dick**, KA Meyer**, Shawn Levy, B Boone, R Stumpf, T Wynne, PV Zimba, DB Gutierrez, and SW Wilhelm (2017). Ecophysiological examination of the Lake Erie *Microcystis* bloom in 2014: linkages between biology and the water supply shutdown of Toledo, Ohio. *Environ. Sci. and Tech.*, 51: 6745-6755.
- Berry, MA, TW Davis, RM Cory, MB Duhaime, TH Johengen, GW Kling, JA Marino, PA Den Uyl*, D Gossiaux, **GJ Dick**, and VJ Denef (2017). Cyanobacterial harmful algal blooms are a biological disturbance to western Lake Erie bacterial communities. *Environ. Microbiol.*: 19: 1149-1162.
- Berry MA, JD White, TW Davis, S Jain, TH Johengen, **GJ Dick**, O Sarnelle, and VJ Denef (2017). Are oligotypes meaningful ecological and phylogenetic units? A case study of *Microcystis* in freshwater lakes. *Frontiers in Microbiology* 8: 365.
- Cory, RM, TW Davis, **GJ Dick**, TJ Johengen, VJ Denef, MA Berry, SE Page, SB Watson, K Yuhas, & GW Kling (2016). Seasonal dynamics in dissolved organic matter, hydrogen peroxide, and cyanobacterial blooms in Lake Erie. *Frontiers in Marine Science* 3: 54.
- Reed, DC**, JA Breier, H Jiang, K Anantharaman*, CA Klausmeier, BM Toner, C Hancock, K Speer, AM Thurnherr, and **GJ Dick** (2015). Predicting the response of the deep-ocean microbiome to geochemical perturbations by hydrothermal vents. *ISME Journal* 9: 1857-1869.
- Reed, DC**, CK Algar, JA Huber, and **GJ Dick** (2014). Gene-centric approach to integrating environmental genomics and biogeochemical models. *PNAS*. 111: 1879-188.

Other selected recent publications

- Dick GJ** (2019). The microbiomes of deep-sea hydrothermal vents: distributed globally, shaped locally. *Nature Reviews Microbiology* (in press).
- Dick GJ**, SG Grim*, and JK Klatt** (2018). Controls on O₂ production in cyanobacterial mats and implications for Earth's oxygenation. *Annual Reviews of Earth and Planetary Sciences* 46: 123-147.
- Anantharaman*, K, JA Breier and **GJ Dick** (2016). Metagenomic resolution of microbial functions in deep-sea hydrothermal plumes across the Eastern Lau Spreading Center. *ISME Journal* 10: 225-239.
- Anantharaman, KA*, MB Duhaime, JA Breier, K Wendt, BM Toner, and **GJ Dick** (2014). Sulfur oxidation genes in diverse deep-sea viruses. *Science* 344: 757-760.

E. SYNERGISTIC ACTIVITIES

Steering Committee, U-M LSA Theme Semester	on the Great Lakes	2019-present
Editorial Board, <i>Environmental Microbiology</i>		2014-present
Associate Editor for molecular geomicrobiology, <i>Geobiology</i>		2014-present
Faculty Advisor, Earth Camp, a summer camp for high school students		2010-present

Craig A. Stow, Biographical Sketch

NOAA Great Lakes Environmental Research Laboratory, Ann Arbor, MI 48108

734-741-2268, craig.stow@noaa.gov

A. Professional Preparation

B.S., Environmental Technology, Cornell University, Ithaca, NY. 1977.

M.S., Marine Sciences, Louisiana State University, Baton Rouge, LA. 1981

Ph.D. Environmental Modeling, Duke University, Durham, NC. 1992

Post-doctoral Researcher, University of Wisconsin, Center for Limnology, 1992–96.

B. Appointments

Senior Scientist, NOAA Great Lakes Environmental Research Laboratory, 2006-present

Associate Professor, University of South Carolina, Dept. Env. Health Sciences, 2003–06.

Visiting Asst. Professor, Duke University, Nicholas School of the Environment, 1996–2003.

LA Department of Environmental Quality, Baton Rouge, LA, 1982–86.

C. Relevant Publications

Web of Science: Publications – 127; Citations – 4,039; H-Index - 37

Weiskerger, C.J., M.D. Rowe, **C.A. Stow**, D. Stuart, T. Johengen. 2018. Application of the Beer-Lambert model to attenuation of photosynthetically active radiation in a shallow, eutrophic lake. *Water Resources Research*, 54: 8952-8962.

Stow, C.A., K. Webster, T. Wagner, N. Lottig, Y. Cha, and P.A. Soranno. 2018. Small values in big data: the continuing need for appropriate metadata. *Ecological Informatics*, 45: 26-30.

Filstrup, C.T., T. Wagner, S.K. Oliver, **C.A. Stow**, K.E. Webster, E.H. Stanley, and J.A. Downing. 2018. Evidence for regional nitrogen stress on lake phytoplankton biomass across large landscape and climate gradients. *Limnology and Oceanography*, 63: S324-S339.

Lottig, N.R., P.N. Tan, T. Wagner, K.C. Cheruvilil, P.A. Soranno, E.H. Stanley, C.E. Scott, **C.A. Stow**, S. Yuan. 2017. Macroscale patterns of synchrony identify complex relationships among spatial and temporal ecosystem drivers. *Ecosphere*, 8(12):e02024.

Oliver, S.K., S.M. Collins, P.A. Soranno, T. Wagner, E.H. Stanley, J.R. Jones, **C.A. Stow**, and N.R. Lottig. Unexpected stasis in a changing world: Lake nutrient and chlorophyll trends since 1990. 2017. *Global Change Biology*, 23: 5455-5467.

Reavie, E.D., G.V. Sgro, L.R. Estep, A.J. Bramburger, V.L. Shaw Chraibi, R.W. Pillsbury, M. Cai, **C.A. Stow**, and A. Dove. 2017. Climate warming and changes in *Cyclotella sensu lato* in the Laurentian Great Lakes. *Limnology and Oceanography*, 62: 768-783.

Nojavan, F., S.S. Qian, and **C.A. Stow**. 2017. Comparative analysis of discretization methods in Bayesian networks. *Environmental Modelling and Software*, 87: 64-71.

Cha, Y., I. Alameddine, S.S. Qian, and **C.A. Stow**. 2016. A cross-scale view of N and P limitation using a Bayesian hierarchical model. *Limnology and Oceanography*, 61: 2276-2285.

Bertani, I., A.D. Gronewold, D. Obenour, C. Steger, **C.A. Stow**, D. Scavia. 2016. Probabilistically assessing the role of nutrient loading in harmful algal bloom formation in western Lake Erie. *Journal of Great Lakes Research*, 42: 1184-1192.

Stow, C.A. 2015. The need for sustained, long-term phosphorus modeling in the Great Lakes. *Journal of Great Lakes Research*, 41: 315-316.

- Stow, C.A.,** Y. Cha, L.T. Johnson, R. Confesor, R.P. Richards. 2015. Long-term and seasonal trend decomposition of Maumee River nutrient inputs to western Lake Erie. *Environmental Science & Technology*, 49: 3392-3400.
- Gronewold, A.D., and **C.A. Stow**. 2014. Water loss from the Great Lakes. *Science*, 343: 1084-1085.
- Stow, C.A.,** J. Jolliff, D. McGillicuddy, S. Doney, J.I. Allen, M. Friedrichs, K. Rose, and P. Wallhead. 2009. Skill assessment for coupled biological/physical models of marine systems. *Journal of Marine Systems*, 76: 4-15.

D. Relevant Recent Grants

2017-2022 Operational Lake Erie Hypoxia Forecasting for Public Water Systems Decision Support. NOAA Center for Sponsored Coastal Ocean Research. \$1,542,314. PI.

E. Synergistic Activities

- International Joint Commission Adaptive Management/Modeling Work Group
- Great Lakes Net Pen Aquaculture Science Panel
- Great Lakes Water Quality Agreement Annex 2 Lake Ecosystems Objectives Task Team
- Great Lakes Water Quality Agreement Annex 4 Nutrient Subcommittee
- Managing for Resilience. Working Group. John Wesley Powell Center for Earth System Science Analysis and Synthesis

John F. Bratton, Ph.D., Biographical Sketch

LimnoTech, 501 Avis Drive, Ann Arbor, MI, 48108
(734) 332-1200, jbratton@limno.com

(a) Professional Preparation

Brown University, Providence, RI	Geology-Chemistry	Sc.B., 1987
University of California at Berkeley	Geology	Ph.D., 1997

(b) Appointments

Academic:

2012-present Wayne State University (MI), Adjunct Faculty Member, Advisory Board Member:
Transformative Research in Urban Sustainability Training (T-RUST)

2006-2010 Stonehill College (MA), Environmental Studies, Part-Time Faculty Member

2001-08, '16 Au Sable Inst. of Environ. Studies (MI), Great Lakes, Associate Professor

2001 Boston University, Dept. of Earth Sciences, Lecturer

2000, '07-08 Bridgewater State College, MA, Earth Sciences Dept., Visiting Assoc. Prof.

1993-1995 Univ. of California-Berkeley, Graduate Student Instructor

Professional:

Licensed Professional Geologist in Utah, Alabama, and Florida

2015-present LimnoTech, Ann Arbor, MI, Senior Scientist

2012-2015 NOAA-Great Lakes Environmental Research Laboratory, Ann Arbor, MI, Deputy
Director, Acting Director, Great Lakes Regional Team Lead

1997-2011 U.S. Geological Survey, Coastal & Marine Geol., Woods Hole, MA, Research Geol.

1996-1997 Berkeley Center for Isotope Geochemistry at Lawrence Berkeley National Laboratory,
Graduate Student Researcher

1994-1995 ERM-West, Inc., Walnut Creek, CA, Senior Environmental Geologist

1991-1993 Balsam Enviro. Consultants, Inc. (now AECOM), Salem, NH, Project Geologist

1988-1991 Roy F. Weston, Inc., Concord (now Manchester), NH, Asst. Project Geologist

(c) Publications

(i) *five recent publications most closely related to the proposed project*

Klump, J.V., **Bratton, J.**, Fermanich, K., Forsythe, P., Harris, H.J., Howe, R.W. and Kaster, J.L.,
2018. Green Bay, Lake Michigan: A proving ground for Great Lakes restoration. *Journal of
Great Lakes Research*, 44(5), pp.825-828.

Wang, J., Kessler, J., Bai, X., Clites, A., Lofgren, B., Assuncao, A., **Bratton, J.**, Chu, P. and
Leshkevich, G., 2018. Decadal Variability of Great Lakes Ice Cover in Response to AMO and
PDO, 1963–2017. *Journal of Climate*, 31(18), pp.7249-7268.

Verhamme, Edward M., Todd M. Redder, Derek A. Schlea, Jeremy Grush, John F. Bratton, and Joseph V. DePinto. Development of the Western Lake Erie Ecosystem Model (WLEEM): Application to connect phosphorus loads to cyanobacteria biomass. *Journal of Great Lakes Research* 42, no. 6 (2016): 1193-1205.

Bratton, J., E. Verhamme, and P. Freedman, 2016, Lessons from Toledo's water crisis about dealing with algal blooms, Proceedings of the Water Environment Federation, WEFTEC 2016, Session 411: Public Health in the Headlines, p.567-568, <https://doi.org/10.2175/193864716819713943>

Baskaran, M., and **J.F. Bratton**, 2013, Investigating human-induced changes of elemental cycles in the Great Lakes, EOS, vol. 94, no. 28, p. 248, DOI:10.1002/eost.v94.28/issuetoc (workshop supported by NSF-Chemical Oceanography Program).

(ii) *five other significant publications*

Gronewold, A.D., E.J. Anderson, B.M. Lofgren, P.D. Blanken, J. Wang, J.P. Smith, T.S. Hunter, G.A. Lang, C.A. Stow, D. Beletsky, and **J.F. Bratton**, 2015, Impacts of extreme 2013-2014 winter conditions on Lake Michigan's fall heat content, surface temperature, and evaporation, *Geophysical Research Letters* 42, 7 pp., DOI:10.1002/2015GL063799.

Szymczycha, B., Kroeger, K.D., Crusius, J., and **Bratton, J.F.**, 2017, Depth of the vadose zone controls aquifer biogeochemical conditions and extent of anthropogenic nitrogen removal, *Water Research*, 123:794-801.

Russoniello, Christopher J., Cristina Fernandez, **John F. Bratton**, Joel F. Banaszak, David E. Krantz, A. Scott Andres, Leonard F. Konikow, and Holly A. Michael, 2013, Geologic effects on groundwater salinity and discharge into an estuary, *Journal of Hydrology*, vol. 498, p. 1-12.

Bratton, John F., 2010, The three scales of submarine groundwater flow and discharge across passive continental margins, *Journal of Geology*, vol. 118, p. 565-575.

Bratton, John F., Böhlke, J.K., Krantz, David E., and Tobias, Craig R., 2009, Flow and geochemistry of groundwater beneath a back-barrier lagoon: The subterranean estuary at Chincoteague Bay, Maryland, USA, *Marine Chemistry*, vol. 113, p. 78-92.

(d) **Synergistic Activities**

2018-19: Developed syllabus for Wayne State Univ. online course on harmful algal blooms

2017-present: LimnoTech project manager for GLOS DMAC support; IOOS Office of Tech Transfer project: Upgrading and Planning for the Transition of the Lake Erie Harmful Algal Blooms Early Warning System to a Sustainable Operational Form; and NOAA-NCCOS ECOHAB project: Linking Process Models and Field Experiments to Forecast Algal Bloom Toxicity In Lake Erie

2013-2017: U.S.-Canada International Joint Commission, Science Advisory Board--Research Coordination Committee; U.S.-Canada Great Lakes Water Quality Agreement Annex 10 (Science) Subcommittee; Steering Committee, NASA/NOAA/MTRI Great Lakes Workshop Series on Remote Sensing of Water Quality; Co-director of Great Lakes Biogeochemistry Initiative, (NSF-funded); Climate Change Executive Forum Panel, Federal Executive Institute (Convener); Great Lakes Restoration Initiative Regional Working Group, Science Subgroup

Edward M. Verhamme, Biographical Sketch
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734-332-1200, everhamme@limno.com

(a) Professional Preparation

Michigan Technological University Houghton, MI Environmental Engineering, M.S., 2005
Michigan Technological University Houghton, MI Environmental Engineering, B.S., 2003
Manhattan College, Yonkers, NY Water Quality Modeling Course 2006

- Modeling of eutrophication, toxic substances, and metals in river and lake systems

(b) Appointments

- June 2005–present, Project Engineer, LimnoTech, Ann Arbor, Michigan
- June 2003–2005, Research Assistant, Michigan Technological University, Houghton, MI
- Summer 2002, Environmental Technician, West U.P. District Health Dept., Hancock, MI
- Summer 2001, Engineering Assistant, City of Escanaba, MI

(c) Publications

(i) five recent publications most closely related to the proposed project

- Verhamme, E.M. et al. 2016. Development of the Western Lake Erie Ecosystem Model (WLEEM): Application to connect phosphorus loads to cyanobacteria biomass. *J. Great Lakes Res.* <http://dx.doi.org/10.1016/j.jglr.2016.09.006>
- Sesterhenn, T.M., C.R. Roswell, S.R. Stein, P. Klaver, **E. Verhamme**, S.A. Pothoven, T.O. Hook. 2014. Modeling the implications of multiple hatching sites for larval dynamics in the resurgent Saginaw Bay walleye population. *J. Great Lakes Res.*, 40(Supp. 1):113-122.
- Van Cleave, K., J.D. Lenters, J. Wang, **E.M. Verhamme**. 2014. A regime shift in Lake Superior ice cover, evaporation, and water temperature following the warm El Niño winter of 1997–1998. *Limnology & Oceanography*, 59:1889-1898.
- Hawley, N., T. Redder, R. Beletsky, **E. Verhamme**, D. Beletsky, J.V. DePinto. 2014. Sediment resuspension in Saginaw Bay. *J. Great Lakes Res.*, 40(Supp. 1):18-27.
- Michalak A.M., ... **E.M. Verhamme**, and 27 others. 2013. Record-setting algal bloom in Lake Erie caused by agricultural and meteorological trends consistent with expected future conditions. *Proc. National Academy of Sci.*, 110:6448-6452.

(ii) five other significant publications

- Bratton, J., **E. Verhamme**, and P. Freedman. 2016. Lessons from Toledo's water crisis about dealing with algal blooms. *Proceedings of the Water Environment Federation*, WEFTEC 2016, Session 411: Public Health in the Headlines, p.567-568, <https://doi.org/10.2175/193864716819713943>
- Kaur, J., J.V. DePinto, J.F. Atkinson, **E. Verhamme**, T.C. Young. 2012. Development of a spatially resolved linked hydrodynamic and exposure model (LOTOX2) for PCBs in Lake Ontario. *J. Great Lakes Res.*, 38:490-503.
- Verhamme, E.M.**, M.T. Auer. 2009. Comparison of carbon flux and indicators of bioavailability at nearshore and offshore locations in Lake Superior. *Verh. Int. Ver. Limnol.* 30:1239–1241.

DePinto, Joseph V., Holmberg, Hans, Redder, Todd, **Verhamme, Edward**, Larson, Wendy, Senjem, Norman J., Munir, Hafiz. 2009. Linked hydrodynamic-sediment transport-water quality model for support of the upper Mississippi River – Lake Pepin TMDL. Proceedings of the Water Environment Federation, TMDL 2009, p. 212-231(20).

Verhamme, E.M. 2005. Comparing Indicators of Bacterial Metabolic Activity at Slope and Profundal Sites in Lake Superior Sediments. Thesis. Dept. of Civil and Environmental Engineering, Mich. Technological University.

(d) Synergistic activities

- Maintain Great Lakes Twitter account (@EddieGreatLakes) with ~1,500 followers, >3,100 tweets, >8,500 “likes”
- Board member: International Association for Great Lakes Research. 2018-present.
- Recent Conference Presentations: Annual Conferences on Great Lakes Research (Univ. of Guelph, Univ. of Vermont, Purdue, McMaster Univ., U-Toronto); Marine Technology Society (Woods Hole Oceanographic Inst.); National Weather Service Forecasters Meeting (Chicago). 2013-2019.
- Classroom and public meeting presentations, mentoring of interns, advising of graduate students and postdocs (Univ. of Mich., Univ. of Wisconsin-Milwaukee, Michigan Tech, Univ. of Toledo).
- Support of City of Toledo Water Intake Monitoring in Lake Erie. 2014-present.
- Support of Lake Study for Offshore Wind Development Project, Lake Erie. 2016.
- Lake Erie HABs and DO Monitoring. 2014-present.
- Western Lake Erie Water Quality Modeling. 2010-2016.
- Development of 3-D Hydrodynamic Water Quality Model of a Calif. Impoundment. 2014-pres.
- Configuration, Assembly, and Deployment of Real-Time Monitoring Buoys in the Great Lakes (n = 12 in 2016). 2011-present.
- Preparation of a Discussion Paper on Lake Erie Phosphorus Targets. 2013.
- Support of Water Quality Model Development in Green Bay, Lake Michigan. 2013-present.
- Development of a Process-Oriented Aquatic Ecosystem Model to Investigate Multi-Stressor Interactions for Saginaw Bay, Lake Huron, MI. 2008-present.
- Development of a Phosphorus Mass Balance Model for Mississquoi Bay, VT. 2011-2012.
- Data Management and Service Support for Great Lakes Observing System. 2011-present.
- National Science Foundation - Lake Erie Biocomplexity Project. 2009.
- Hydrodynamic and Water Quality Modeling of Sandusky Bay, Lake Erie. 2009.

9. Ongoing support

GREGORY J. DICK

Current:

Project/Proposal Title: *GP-IMPACT: Broadening pathways to geosciences with an integrated program at The University of Michigan*

Source of Support: NSF

Point of Contact: Jill Karsten, 703-292-8500, jkarsten@nsf.gov

Project Location: University of Michigan

Total Award Amount: \$226,033

Starting Date: 09/01/15

Ending Date: 8/31/18

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.50

Role: PI

Project/Proposal Title: *Revealing the interplay between light, sulfur cycling, and oxygen production in cyanobacterial mats*

Source of Support: NSF Geobiology and Low-Temperature Geochemistry.

Point of Contact: Hailiang Dong, hdong@nsf.gov

Project Location: University of Michigan

Total Award Amount: \$387,151

Starting Date: 7/01/16

Ending Date: 6/30/19

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.50

Role: PI

Project/Proposal Title: *The role of heterotrophic bacteria in protecting cyanobacteria from hydrogen peroxide in coastal systems*

Source of Support: National Science Foundation

Project Location: University of Michigan

Total Award Amount: \$874,085

Starting Date: 01/01/18

Ending Date: 12/31/20

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.50

Role: PI

****This project is complementary to the current proposal**

Project/Proposal Title: *Development of a gene-based model of toxin production by Microcystis aeruginosa in Lake Erie*

Source of Support: NOAA/Cooperative Institute for Great Lakes Research

Project Location: University of Michigan

Total Award Amount: \$70,000

Starting Date: 06/01/18

Ending Date: 5/31/19

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.50

Role: PI

****This project is complementary to the current proposal**

Project/Proposal Title: *Discovery and characterization of new toxins in genomes and metagenomes of bloom-forming cyanobacteria*

Source of Support: National Institute of Environmental Health Sciences (Prime) – Bowling Green State University (Direct)

Project Location: University of Michigan

Total Award Amount: \$479,519

Starting Date: 10/01/18

Ending Date: 9/30/23

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.50

Role: PI

****This project is complementary to the current proposal**

Project/Proposal Title: *Metagenomic insights into metabolic pathways and microbial interactions of the scalp microbiome*

Source of Support: Procter & Gamble

Project Location: University of Michigan

Total Award Amount: \$70,066

Starting Date: 6/15/18

Ending Date: 9/1/19

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.25

Role: PI

Project/Proposal Title: *ECOHAB 2017: Linking process models and field experiments to forecast algal bloom toxicity in Lake Erie.*

Source of Support: NOAA (Prime), Ohio State University (Direct)

Project Location: University of Michigan

Total Award Amount: \$51,293

Starting Date: 9/1/18

Ending Date: 8/31/20

Support Type: Current

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.00

Role: PI

Pending:

Project/Proposal Title: *P01: Host and Microbial Metabolism in Graft versus Host Disease*

Source of Support: National Institute of Health

Project Location: University of Michigan

Total Award Amount: \$1,014,000

Starting Date: 9/01/19

Ending Date: 8/31/24

Support Type: Pending

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.60

Role: PI

Project/Proposal Title: *Great Lakes Environmental Research Laboratory FY 2019 'Omics Workplan*

Source of Support: NOAA

Project Location: University of Michigan

Total Award Amount: \$700,000 (\$81,429 to GD)

Starting Date: 7/01/19

Ending Date: 6/30/20

Support Type: Pending

Person-months Per Year Committed to the Project: Cal: 0.00 Acad: 0.00 Sumr: 0.00

Role: Co-PI

****This project is complementary to the current proposal**

10. Data Sharing Plan

This project will conform to NOAA's Data Sharing Directive for Grants, Cooperative Agreements, and Contracts. In particular, all data will be made publicly available no later than publication of a peer-reviewed article, or two years after the original end date of the grant.

1. Types of samples, data, and other materials to be produced in the course of the project.

This project will generate the following types of samples, data, and products collected from May 2020 to January 2022:

- Samples: Isolates of *Microcystis*. DNA extracts will be produced from culturing samples and field samples from Lake Erie.
- Data: Nucleic acid sequencing from *Microcystis* isolates and field samples, various physiological traits, mainly Monod equation parameters. LC-MS/MS data from metabolite characterization.
- Products: In addition to peer-reviewed journal papers, we will generate bioinformatic code, written in R, ruby, and python that will be used to process the raw sequence data and perform statistical analyses. Further, a database of physiological traits associated with each *Microcystis* isolate will be generated.

2. Data Quality.

Sample collectors and data generators will focus on standard and customary QA/QC protocols for the methods they are using to collect and analyze samples. Accuracy, precision, completeness, representativeness, comparability, and detection limits will be considered in evaluating data quality.

Genetic data quality will be assured by removing replicated reads (i.e., 100% identical reads with same start, same stop, same sequence) due to artifacts during the sequencing reaction. Low quality sequencing reads (containing unidentified bases) will be removed and the tail ends of reads that do not pass established quality thresholds will be trimmed prior to assembly and gene calling.

Traits values quality will be assured by replication of each culture condition in triplicate and assessing that the coefficient of variance is <20%.

3. Methods and policies for providing access and enabling sharing.

Data collected and products produced for this project will be fully and openly shared with the public.

Genetic data. For public release, raw sequencing data sets (in fastq format) will be deposited in the NCBI Trace Archive (<http://www.ncbi.nlm.nih.gov/Traces/home/>). We will also submit metadata (e.g., water source, sampling, nucleic acid extraction, and sequencing procedures) using the MIMS (Minimum Information About a MetaGenome Sequence) genome metadata package as part of the NCBI trace archive submission process.

Computer code: All our informatic code, including modeling, data processing, and figure generation code will be made available as github repositories on our lab's github page (<https://github.com/Geo-omics/>).

Traits data: All trait values will be stored in a database, which will be made available as metadata to the submitted genome sequences and on the lab github page, the lab data page, as well as in the supplemental information associated with published papers.

4. Methods for archiving and preserving access to data and materials.

Cultures characterized as part of this study will be submitted to two culture collections: the Pasteur Culture Collection and the American Type Culture Collection. All microbial filters and derived nucleic acids and proteins will be stored in the -80 freezers of the PI or co-PIs labs for at least 10 years after the completion of the project. All bioinformatic code, raw data, and processed data will be shared when publications presenting the data are published. Data will be stored indefinitely on the NCBI and GitHub data repositories and National Centers for Environmental Information. Prior to public release, all data will be stored on both a physical backup drive on which sequencing and mass spectrometry data were delivered by the sequencing and MS cores, and on the Flux environment data storage service. Significant research results will be written up and submitted for publication.

Sea Grant Data Management Plan Form

Proposal Submission Phase

Title of the Proposal (required answer):

Name of lead PI (required answer): Sea Grant requires that the lead PI serve as the data steward.

Contact Information (required answer):

Dataset Description(s) (required answer): What data will the dataset(s) contain? This includes descriptive details on data types, inclusion of metadata, data format(s), collection times / date ranges, etc. What name(s), if any, will be designated to the dataset(s)?

Do you agree to release all data no later than 2 years after the end-date of the project? (required answer):

Yes

No

Issues (required answer): Are there any legal, access, retention, etc. issues anticipated for the dataset? If yes, please explain.

Data Size: What will be the estimated size of the dataset? Please report estimated number of MB, GB, TB, etc., collected.

Data Format: What format will the dataset utilize? (i.e., Excel file, model code, audio/video recording, etc.)

Ownership (required answer): Who will own the dataset, if not the lead PI?

Post-Processing: What post-processing, QA/QC will this dataset undergo? Who will be responsible for performing this post-processing and QA/QC to prepare the dataset for its deposition into a repository?

Preservation Plan (required answer): What data repositories will be used to host the dataset? If none, how will the data be preserved?

Products: Will any information or data products be developed from this dataset? How will the related costs be supported? Which organization(s) will be producing these products?

Other Comments: Are there any additional comments related to the data that will results from your Sea Grant-funded study?

11. Supporting Documentation

Three letters of support are included below:

1. Dr. Bradley Cardinale, Director of the Cooperative Institute for Great Lakes Research.
2. Dr. George Bullerjahn, Director of the NIEHS/NSF Great Lakes Center for Fresh Waters and Human Health
3. Dr. Justin Chaffin, Senior Researcher and Research Coordinator, Stone Laboratory.

May 22nd, 2019

Re: Letter of support for proposal

Dear Colleagues,

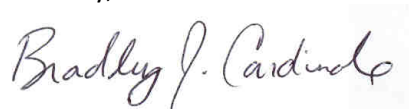
As Director of the Cooperative Institute of Great Lakes Research (CIGLR), I am writing to offer my full support for Dr. Greg Dick's proposal to Michigan Sea Grant to study the ecophysiology of different *Microcystis* strains that cause harmful cyanobacterial blooms in Lake Erie.

In this proposal, Dr. Greg Dick aims to study the combined effect of light, oxidative stress, and nitrogen availability on the growth of different strains of the bloom forming *Microcystis*. The proposed work is an important step towards understanding the triggers of these nuisance blooms. The work builds on, and leverages a considerable body of expertise and data that have been developed from ongoing interactions between Greg Dick's research group, CIGLR, and the NOAA Great Lakes Environmental Research Lab (NOAA-GLERL). CIGLR and NOAA-GLERL have led monitoring efforts of cyanobacterial blooms in Lake Erie for more than a decade, and have developed the Harmful Algal Bloom Tracker that warns water intake managers, public health officials, and charter boat captains of the formation and movement of blooms. Dr. Greg Dick's research group has been part of the weekly CIGLR-NOAA GLERL cruises that monitor cyanobacterial blooms since 2014, and his research group has infused valuable tools and expertise from genomics to help better track and identify toxic strains of cyanobacteria, and to understand the genetic triggers of toxicity.

His newly proposed work to define the key physiological parameters of a different *Microcystis* strains, and to link those traits to genetic markers, is synergistic with NOAA-GLERL and CIGLRs priorities to advance environmental omics methods for the purposes of next generation monitoring and development of next-generation models that aim to achieve predictive capabilities. The data produced by this project, which will be made publicly available, will be valuable to several ongoing efforts by NOAA-GLERL, CIGLR, and partners (e.g., Limnotech) to improve ecological models that can forecast the frequency and severity of blooms, and to predict their movements so that stakeholders can be warned in advance.

I offer my full support for Dr. Greg Dicks project, and will continue to make all of our resources available to his research group if his proposal is funded.

Sincerely,



Brad Cardinale, Professor and Director
Cooperative Institute of Great Lakes Research
School for Environment and Sustainability
University of Michigan
Ann Arbor, MI 48109-1041



George S. Bullerjahn, Ph.D.
Professor and Director
516 Life Sciences Building
Bowling Green State University
Bowling Green OH 43403 USA
(419) 372-8527
bullerj@bgsu.edu
<https://www.bgsu.edu/lake-erie-center.html>

May 16, 2019

Gregory J. Dick, Ph.D.
Associate Professor
Department of Earth & Environmental Sciences
University of Michigan
2534 1100 North University Building
Ann Arbor, MI 48109-1005

Dear Greg,

This letter is in support of your proposal to the Michigan Sea Grant “Determining how the ecophysiology of different *Microcystis* strains underpins succession and toxicity of harmful cyanobacterial blooms in Lake Erie”.

As Director of the recently funded NIEHS/NSF Great Lakes Center for Fresh Waters and Human Health, I am interested in extending further collaborative interactions among Center personnel such as yourself. The Center is a large, multi-institutional program that seeks to characterize cyanobacterial toxins and the environmental conditions that influence their production in the Great Lakes. Through both our NIEHS/NSF funding and existing collaborations with you and other Center members, the Center has substantial resources and infrastructure for sampling, DNA sequencing, research, and public outreach, and I am delighted to leverage these resources to amplify the results and impacts of your proposed research.

I was also pleased to hear of your success in culturing numerous new strains of *Microcystis* from Lake Erie, and of your plans to characterize these strains in the proposed Sea Grant project. Our understanding of the physiological traits of diverse *Microcystis* strains from Lake Erie is not sufficient, yet this knowledge is crucial for understanding and predicting the dynamics of toxin production through the season in Lake Erie. Your proposed work in characterizing the combined effects of light, oxidative stress, and nitrogen availability on growth the *Microcystis*, and linking these traits from lab cultures to dynamics of field cultures, is absolutely synergistic with the culture and whole-community-based approaches that our Center is taking.

Best wishes with your proposal and I look forward to working with you.

Sincerely yours,

George S. Bullerjahn, Ph.D.
Distinguished Research Professor



May 16, 2019

Dear Greg,

I'm writing to express support for your proposal to the Michigan Sea Grant, entitled "Determining how the ecophysiology of different *Microcystis* strains underpins succession and toxicity of harmful cyanobacterial blooms in Lake Erie".

The proposed work, which seeks to define key physiological parameters of a diversity of *Microcystis* strains, and to link those traits to genetic markers, is synergistic with a NOAA ECOHAB Grant on which I am PI, entitled "ECOHAB 2017 Linking process models and field experiments to forecast algal bloom toxicity in Lake Erie". My project involves synthesizing historical data, conducting experiments, and incorporating results into numerical models of ecological and physical processes to develop hindcasting, nowcasting, and forecasting of harmful algal blooms.

However, our research is focused at the community level, and does not resolve the traits of specific strains of *Microcystis*. The physiology of diverse Lake Erie *Microcystis* strains remains a substantial knowledge gap, and the work you propose will be valuable in developing next generation models that are able to integrate biological knowledge of these particular organisms.

In this context, we are pleased that our ECOHAB grant has received supplemental funding to support you to compare environmental metagenomics data to results from the WLEEM model, as modified through your NOAA CIGLR Fellowship, in order to move us towards strain-resolved ecological models of Lake Erie HABs blooms. The physiological work you propose will be critical in parameterizing these models.

In summary, your project will leverage results and active research associated with my NOAA ECOHAB grant. Further, the results of your project will be valuable to our collaborative work and to the HABs community at large.

Sincerely,

Justin Chaffin