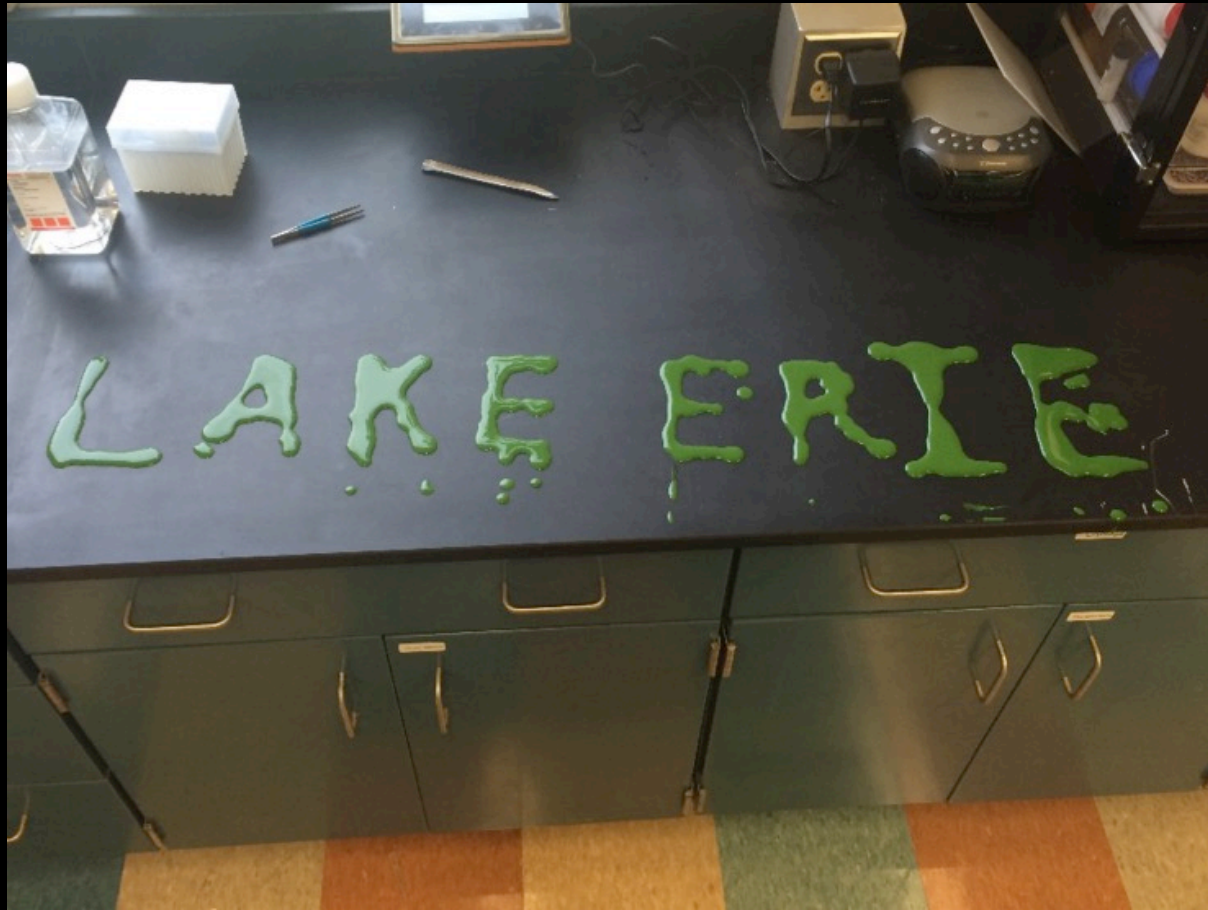
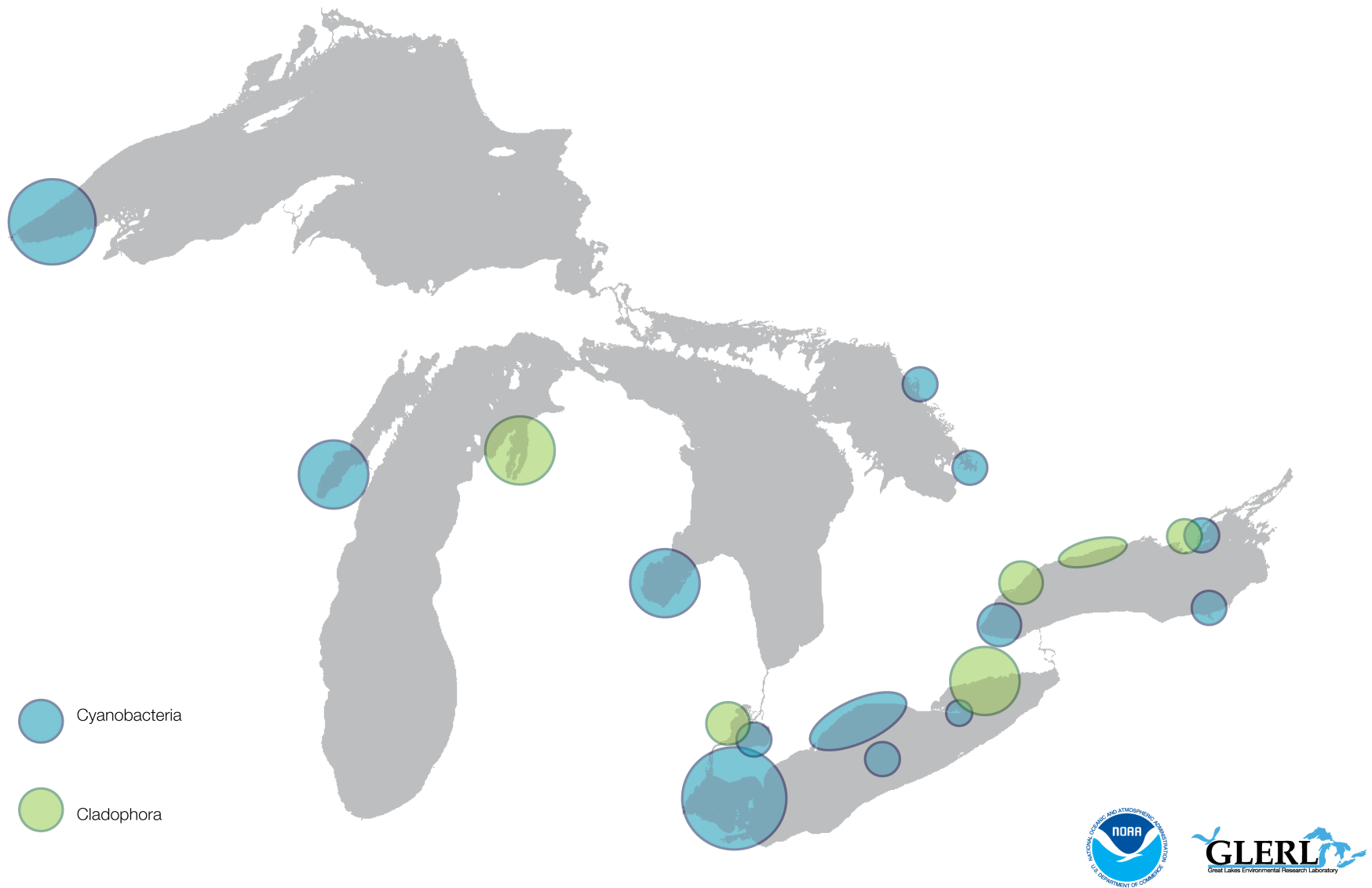


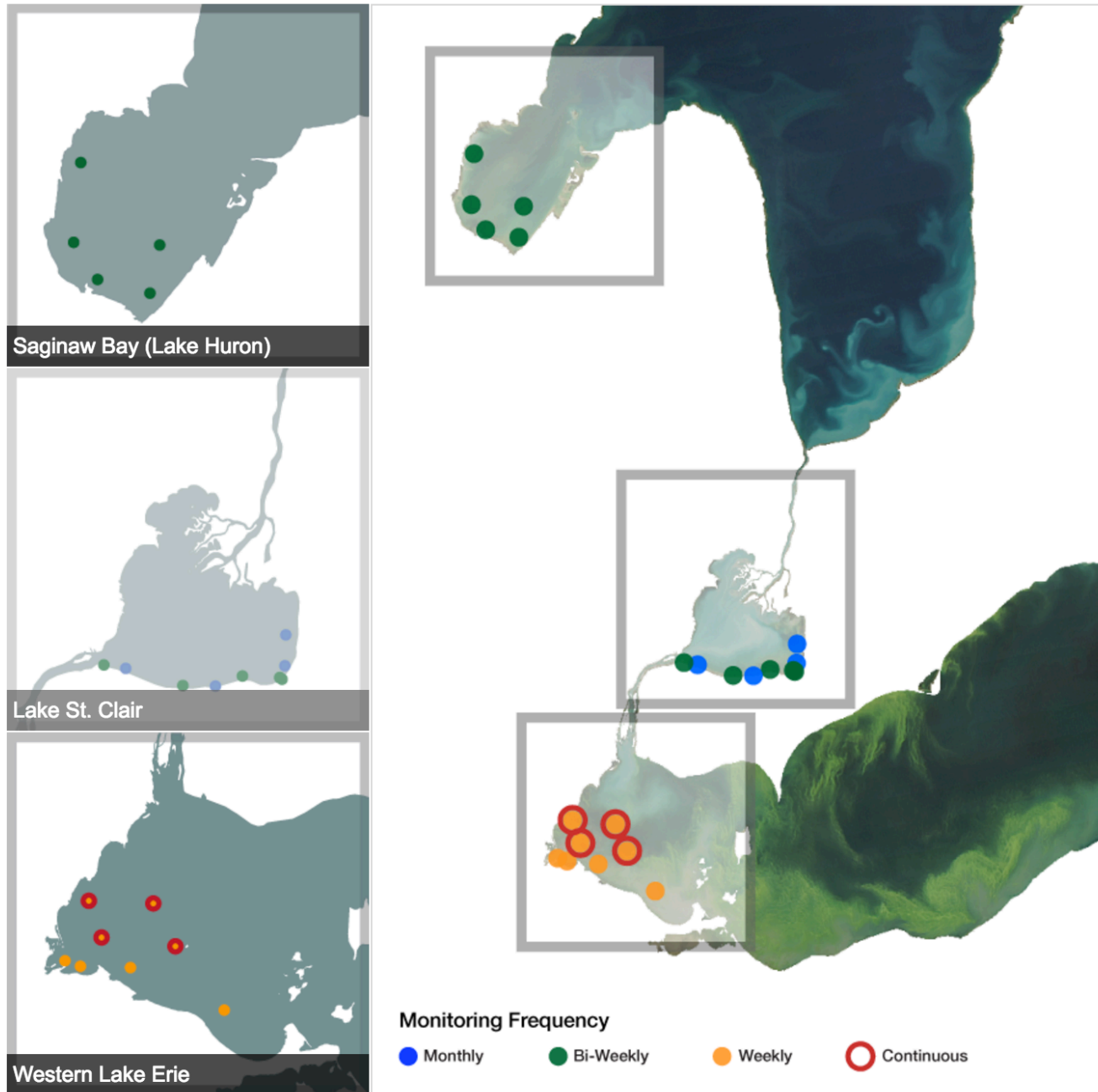
Integrating HAB detection technologies with a regional observing system in the Great Lakes



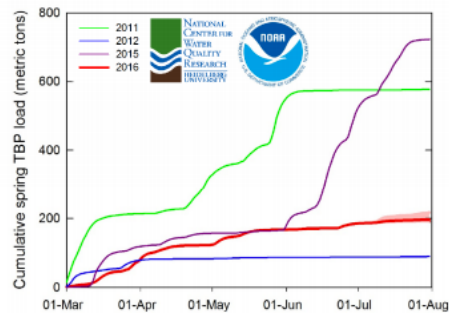
HABs occur throughout the Great Lakes basin



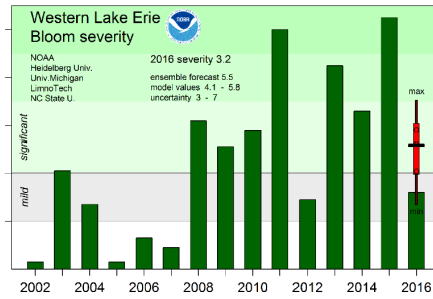
NOAA studies HABs throughout the Great Lakes



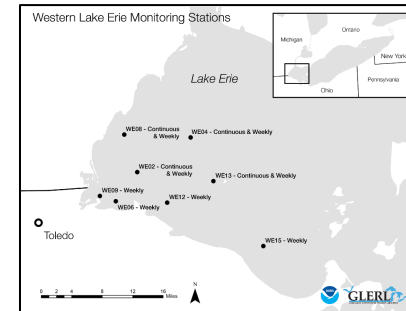
Bloom projection to data dissemination requires a multifaceted effort



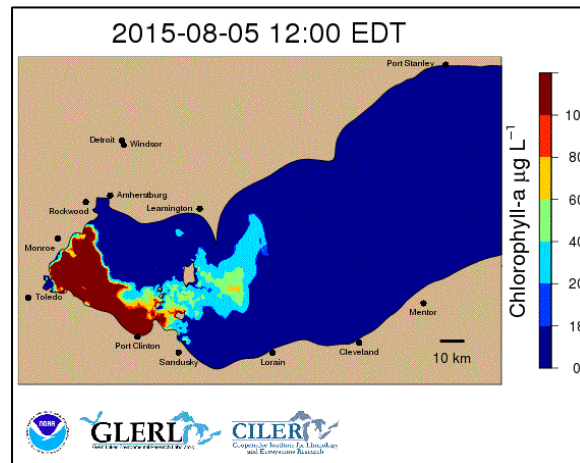
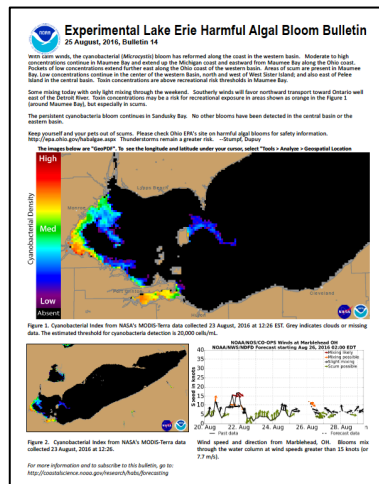
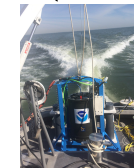
1) Project



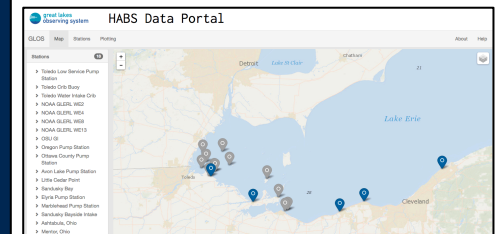
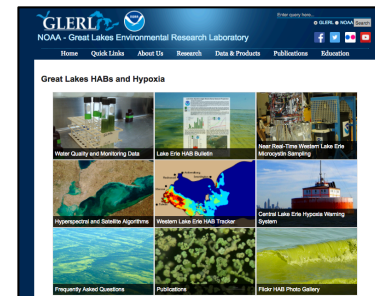
2) Predict



3) Monitor



4) Forecast



5) Disseminate Monitoring Data

NOAA's cross-line office CHAB team

Monitoring & Experiments

Timothy Davis

Tom Johengen*

Greg Doucette

Duane Gossiaux

Ashley Burtner*

Danna Palladino*

Heidi Purcell*

Hank Vanderploeg

Alicia Ritzenthaler*

Remote Sensing

Steve Ruberg

Richard Stumpf

Andrea Vander Woude*

Steve Constant

George Leshkevich

Ron Muzzi

Russ Miller

Modeling

Richard Stumpf

Eric Anderson

Craig Stow

Tim Wynne

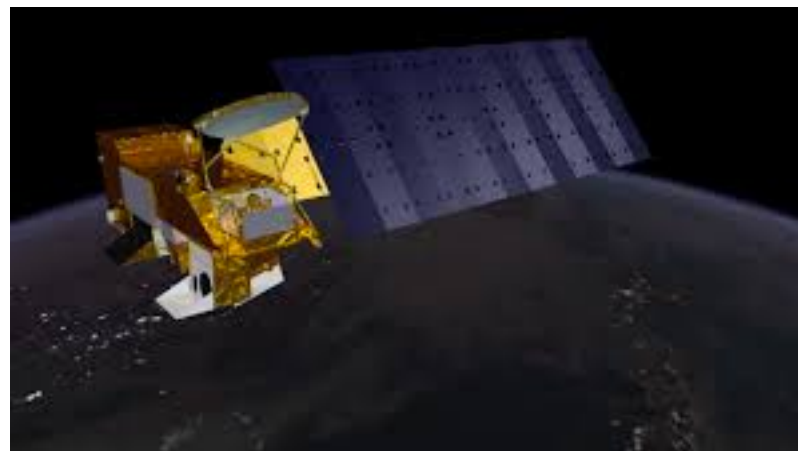
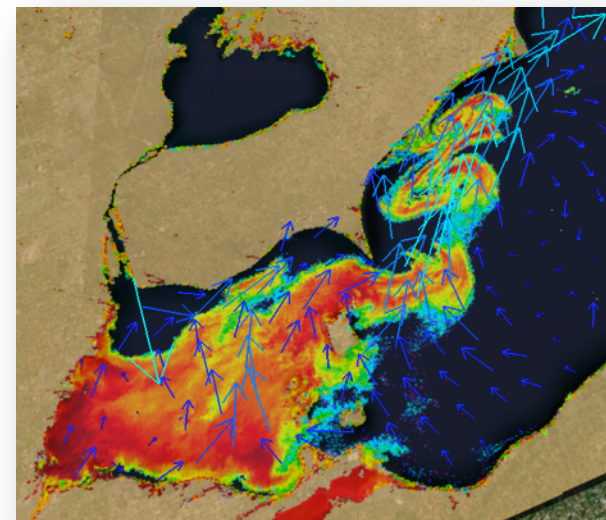
Danielle Dupuy

Mark Rowe*

Greg Lang

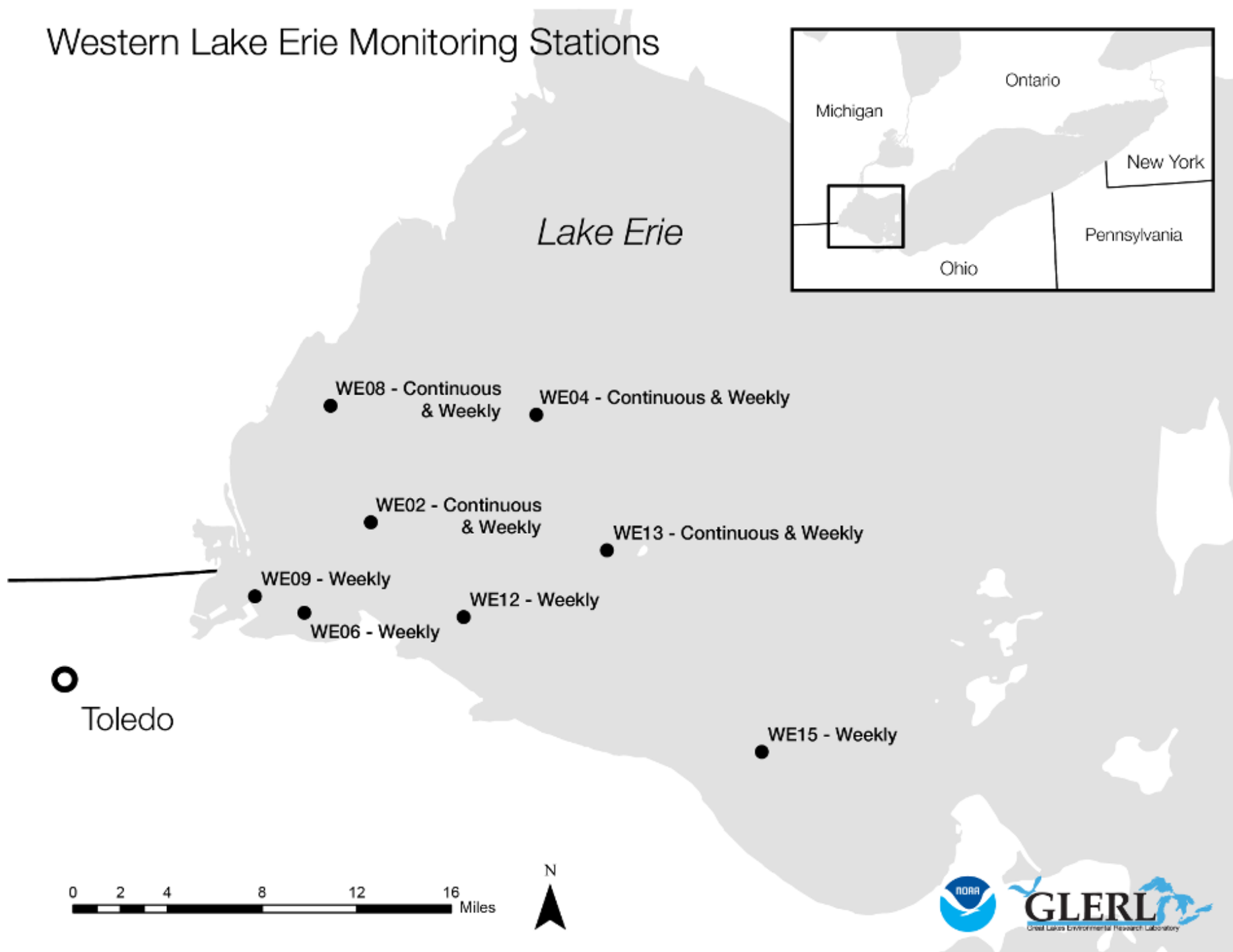
Communications

Joe Smith*



BOLD = HAB team leads
*** = CILER employees**

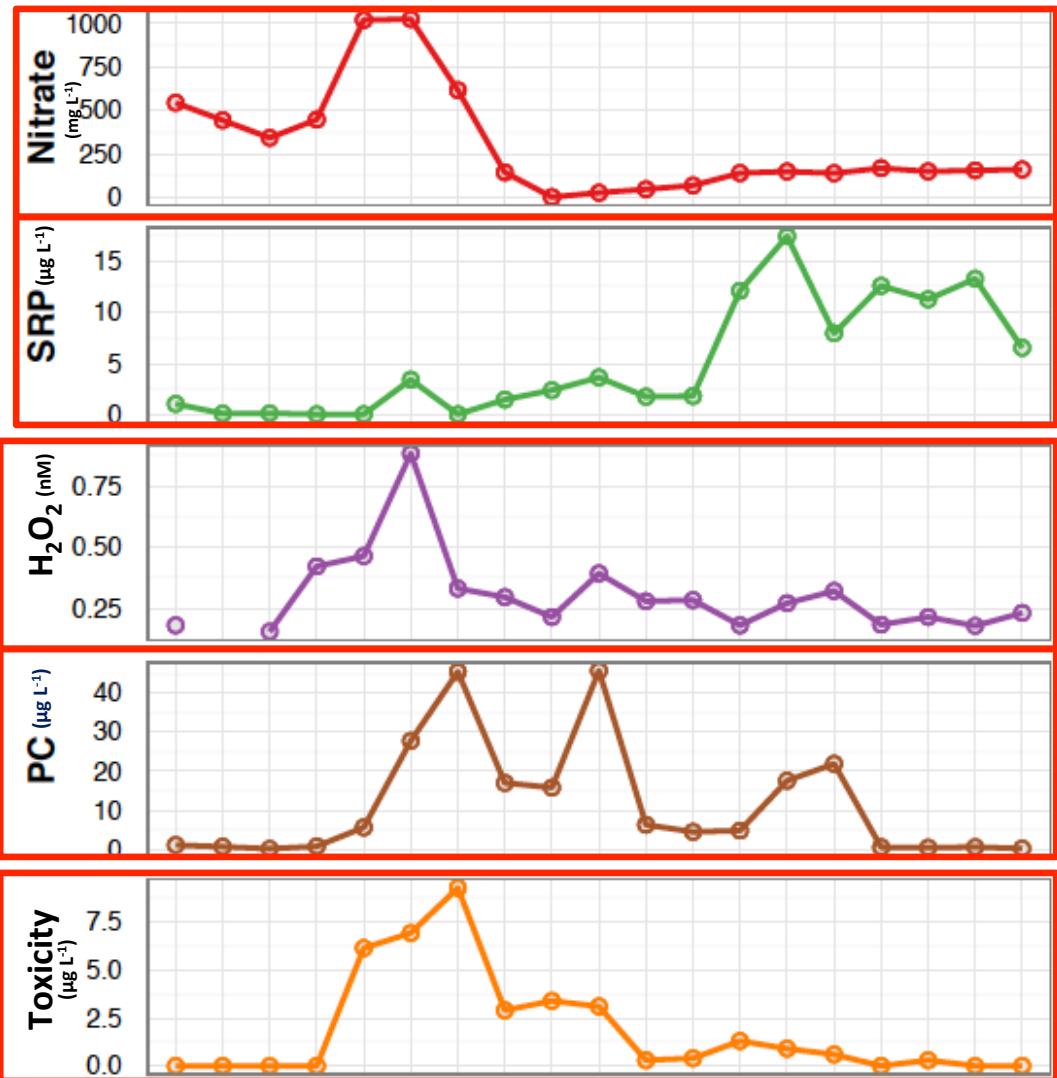
Western Lake Erie Monitoring Stations



Weekly sampling reveals important seasonal trends that highlight potential environmental drivers and inform future experiments

2014 Toledo Water Intake (Station 12)

- Toxicity changes throughout the bloom
- Relationship between nitrogen and toxicity
- *Microcystis* blooms occur even when phosphorus concentrations are low
- Other factors beyond nutrients may be important in driving bloom structure and function



Blooms are patchy!



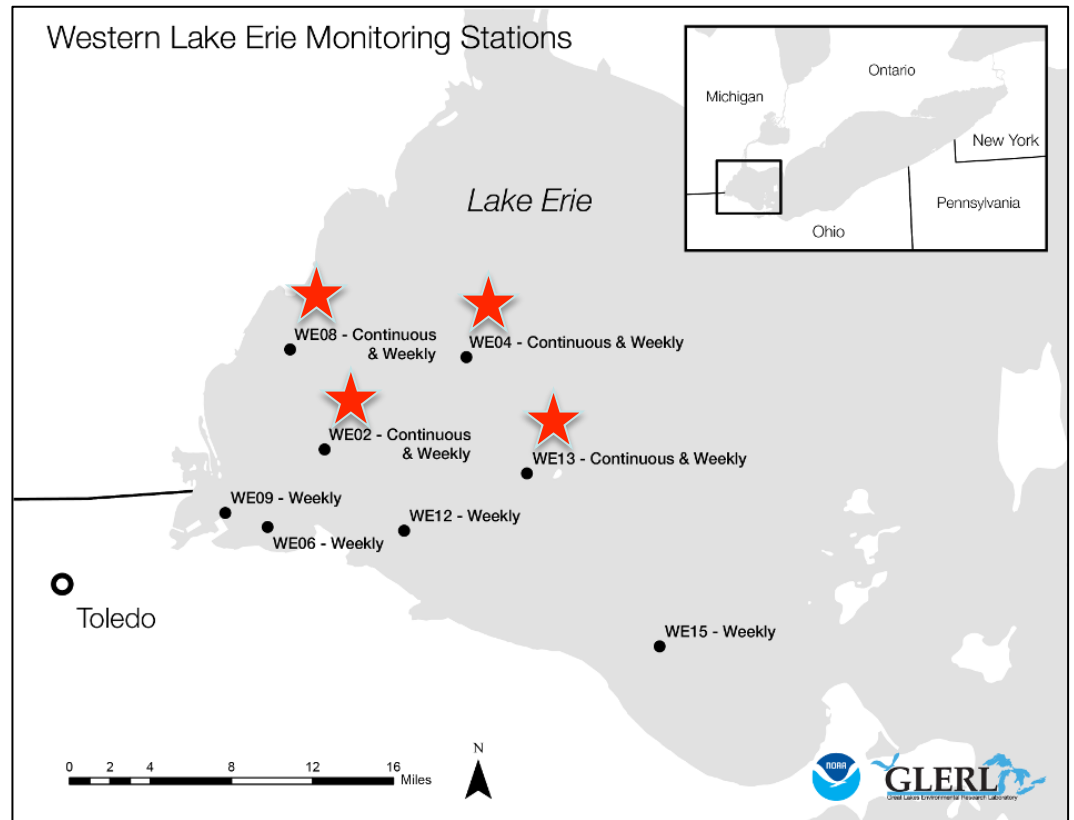
Western Lake Erie Real-time HABs Monitoring

Sensors:

- . YSI EXO sondes
- . Chlorophyll, Phycocyanin, Turbidity, CDOM



- . Wetlabs Cycle -SRP



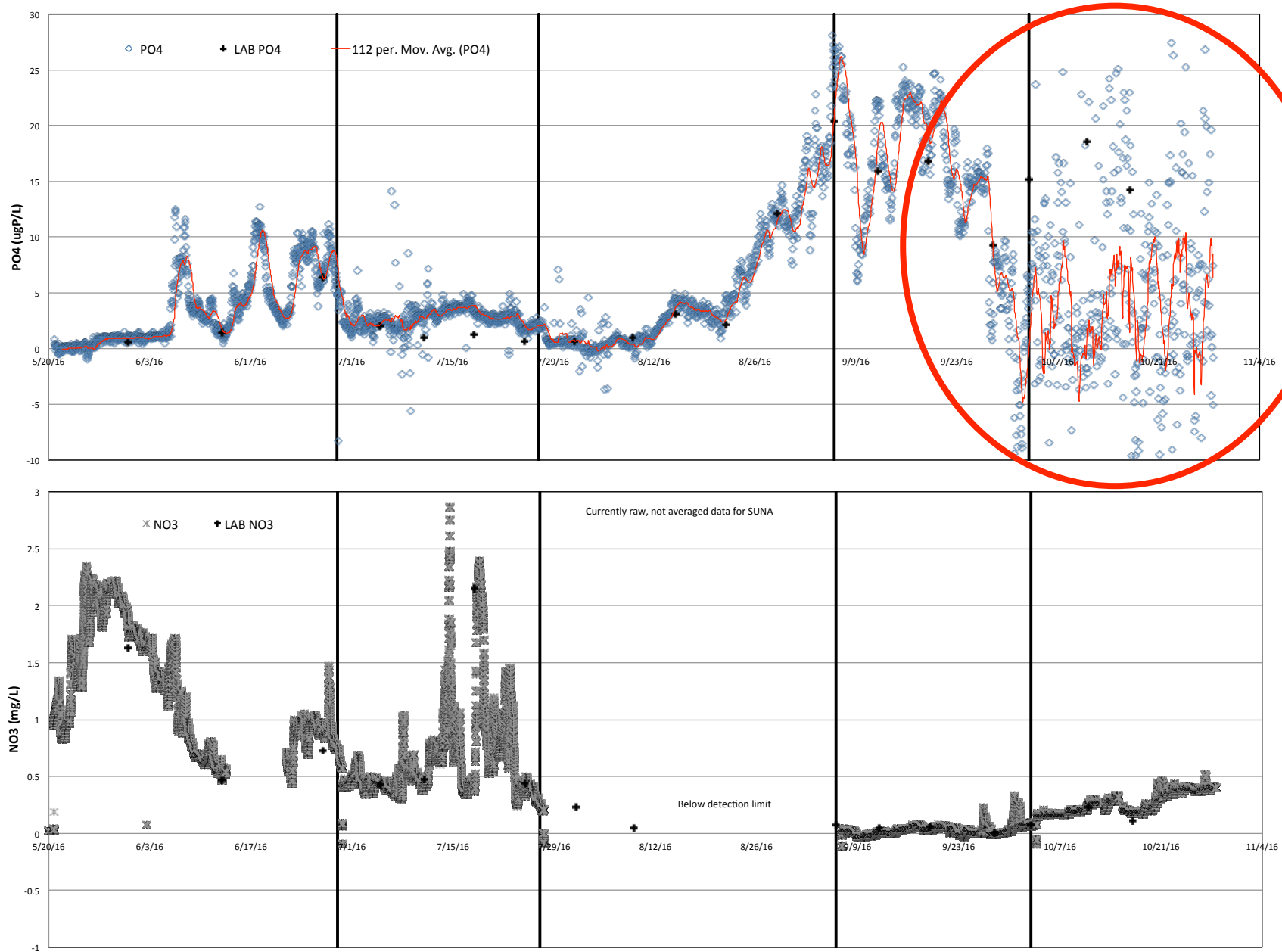
★ = Real-time observation station in 2016

- . SeaBird SUNA nitrate sensor



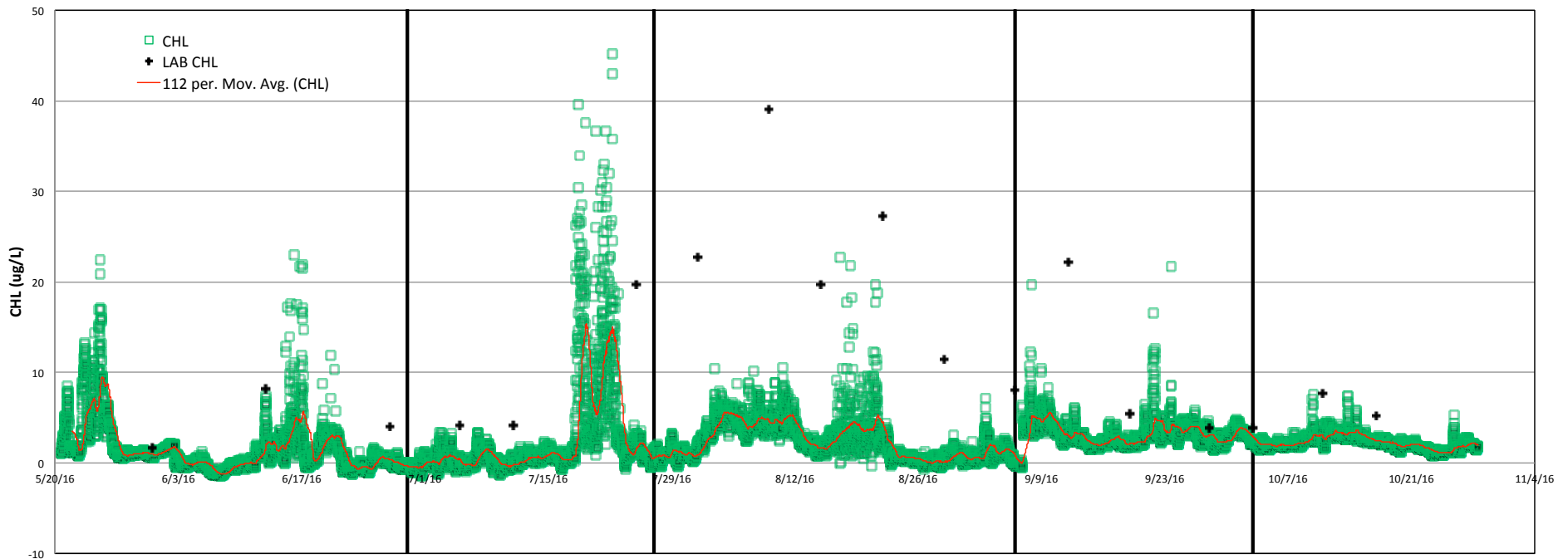
Continuous nutrient monitoring tracks field collections well

Continuous sensor work being conducted by Tom Johengen, Steve Ruberg, Danna Palladino, Heidi Purcell, Steve Constant, Ron Muzzi and Russ Miler



Continuous pigment monitoring yielded more variability

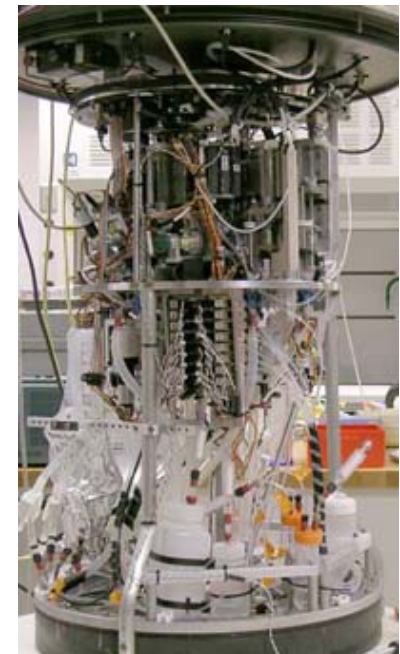
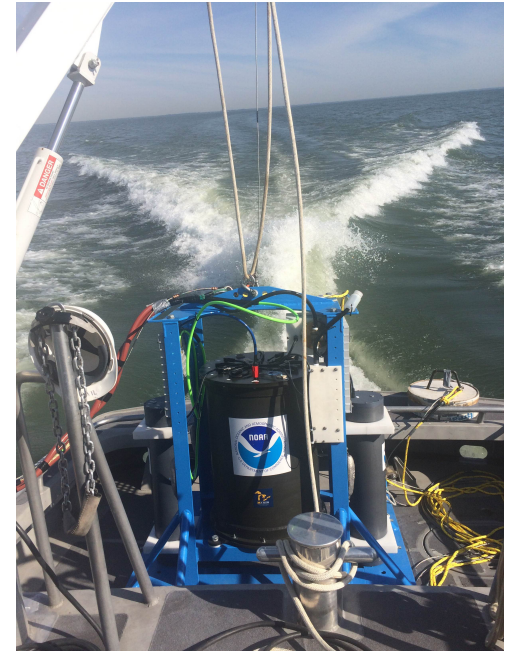
Continuous sensor work being conducted by Tom Johengen, Steve Ruberg, Danna Palladino, Heidi Purcell, Steve Constant, Ron Muzzi and Russ Miler



- Overall trend is seen but much greater variation between grab and continuous data than the nutrient sensors
- ‘Packaging effects’ of colonies interferes with the ability to accurately detect fluorescence signal
- Buoy setup instruments record data about 2m below the surface whereas Niskin samples are taken at 1m
- Highlights the need to be careful when integrating datasets

Autonomous near real-time toxin detection for Lake Erie

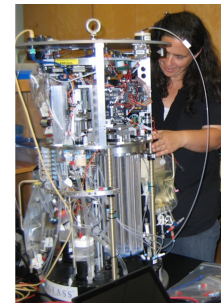
- Purchased with GLRI funds post-Toledo water crisis
- Collaboration between NOAA, MBARI, WHOI, OSU
- Truly emerging technology
 - Fewer than 20 worldwide
 - ESPniagara is the first to study freshwater CHABs
 - **Fine-scale MC observations are critical for toxicity models**
Stumpf, Davis et al., 2016; Harmful Algae



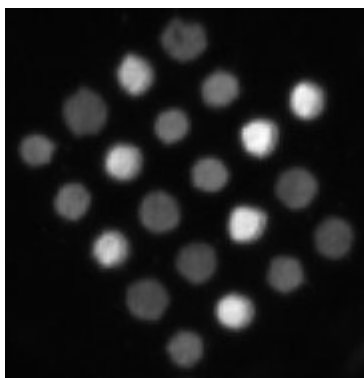


ESP microcystins assay design

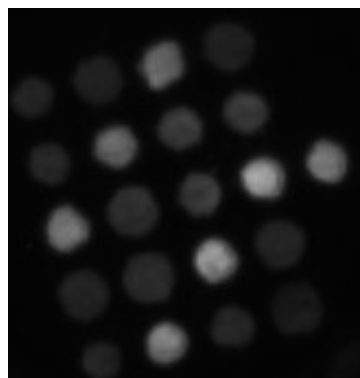
Work being conducted by Greg Doucette and Tina Mikulski



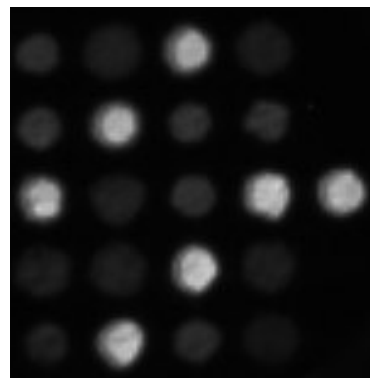
- Using mAb against ADDA moiety and MC-BSA conjugate – for comparison with Abraxis ELISA
- Compatible with extraction solvent
- Good range of detection on benchtop “mimic”
- Good overall intensity values



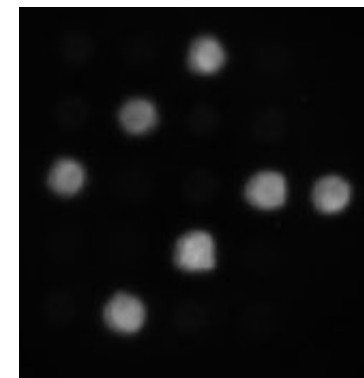
0.2 ng/mL MCLR



2.0 ng/mL MCLR



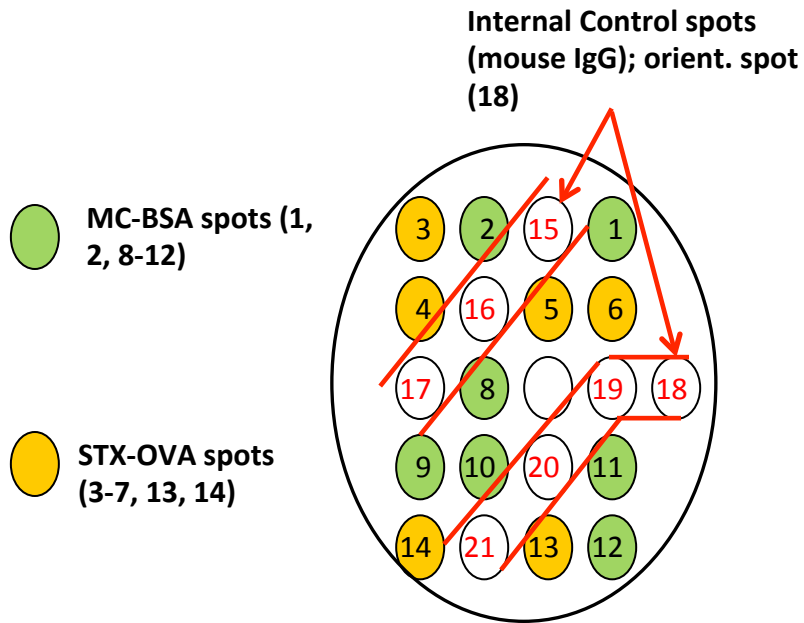
20.0 ng/mL MCLR



200.0 ng/mL MCLR

Potential for multiplexed detection of MC & STX

Work being conducted by Greg Doucette and Tina Mikulski



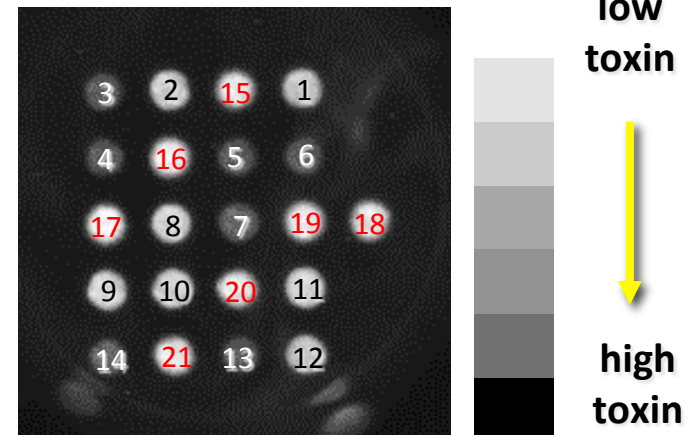
Proposed array map for MC + STX assay
using α -MC & α -STX MAb

example of analogous, multiplexed PST
array for simultaneous detection of
congener groups without (e.g., STX) and
with (e.g., NEO) N-1 OH moiety

- competitive (c) ELISA membrane-based array employs MCLR-BSA & STX-OVA spots
- concurrent application of α -MC and α -STX MAbs in 'cocktail' yields two assay results generated simultaneously on the same array
- will broaden ESP cELISA detection capabilities to include MC, CYL, STX for single sample
- requires development of efficient extraction protocol for MC, CYL, and STX

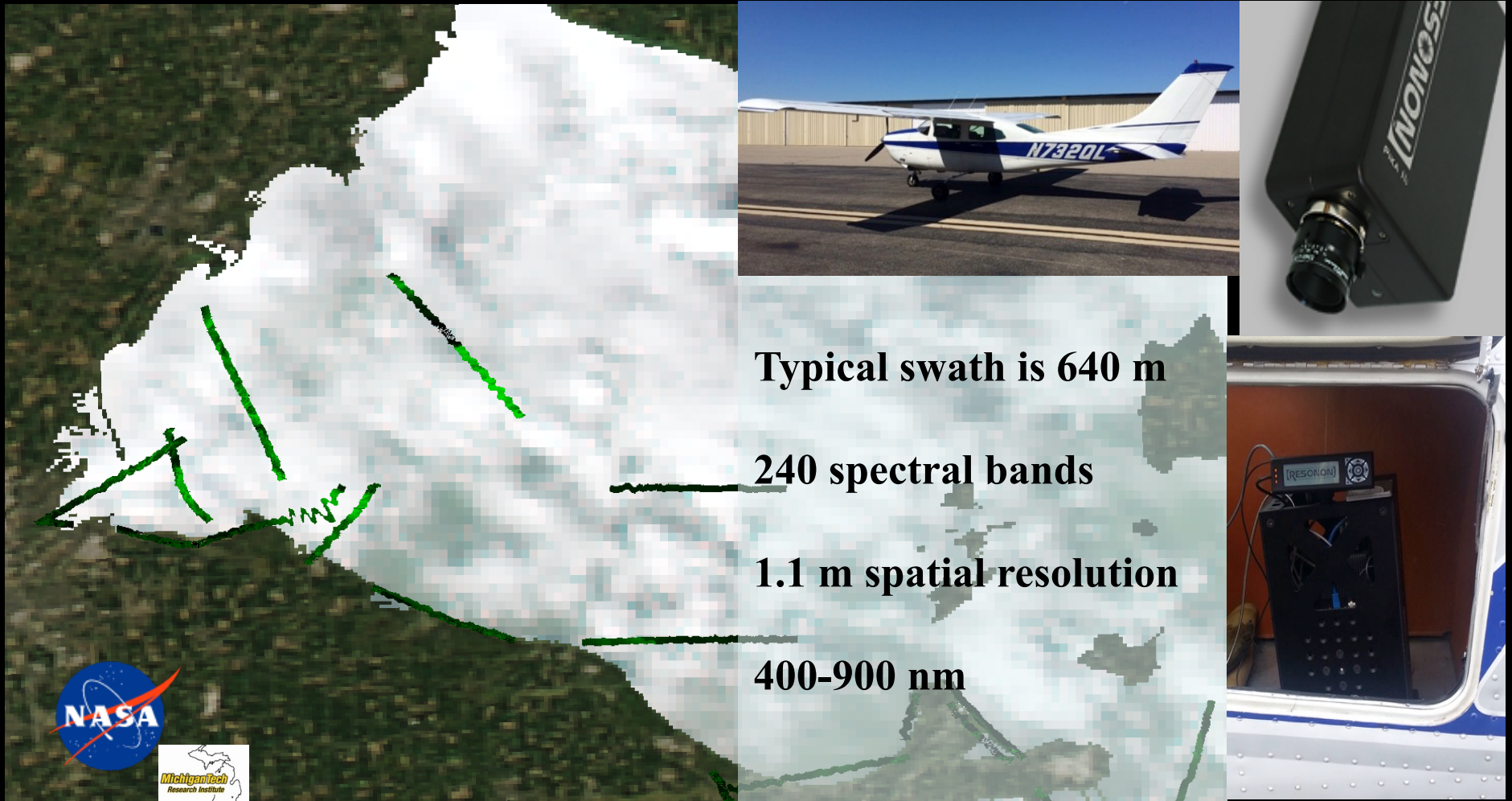
STX-OVA
spots (1, 2,
8-12)

NEO-OVA spots
(3-7, 13, 14)

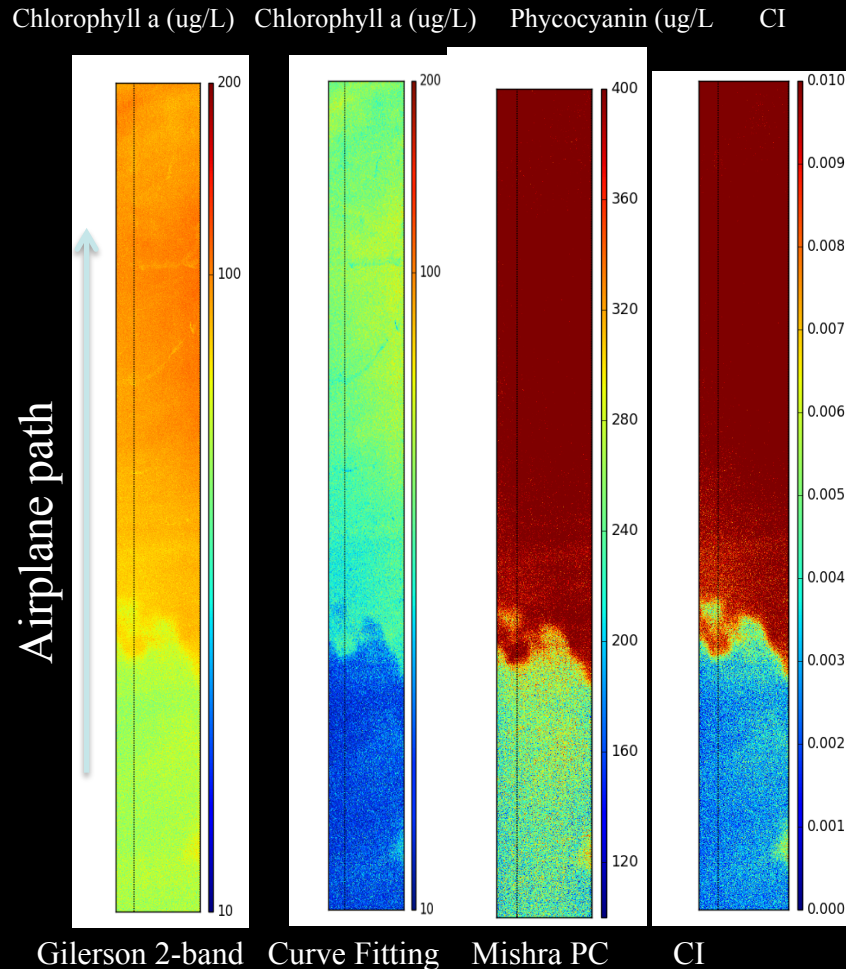


Start of Hyperspectral Detection of Cyanobacteria: Data in small areas; plane and UAS systems.

Lead by Andrea VanderWoude and Steve Ruberg



Algorithm Development; and potential match to satellite to fit into bulletin in the future



(*Images are along track)



August 17, 2015

Recent Scum Event – South of Monroe Water Intake at WE8 9-19-2016 (Truecolor)

Hyperspectral flight found scum
near WE8, 5 km south of Monroe
on Sep-19 at 11:30 AM

Google Earth

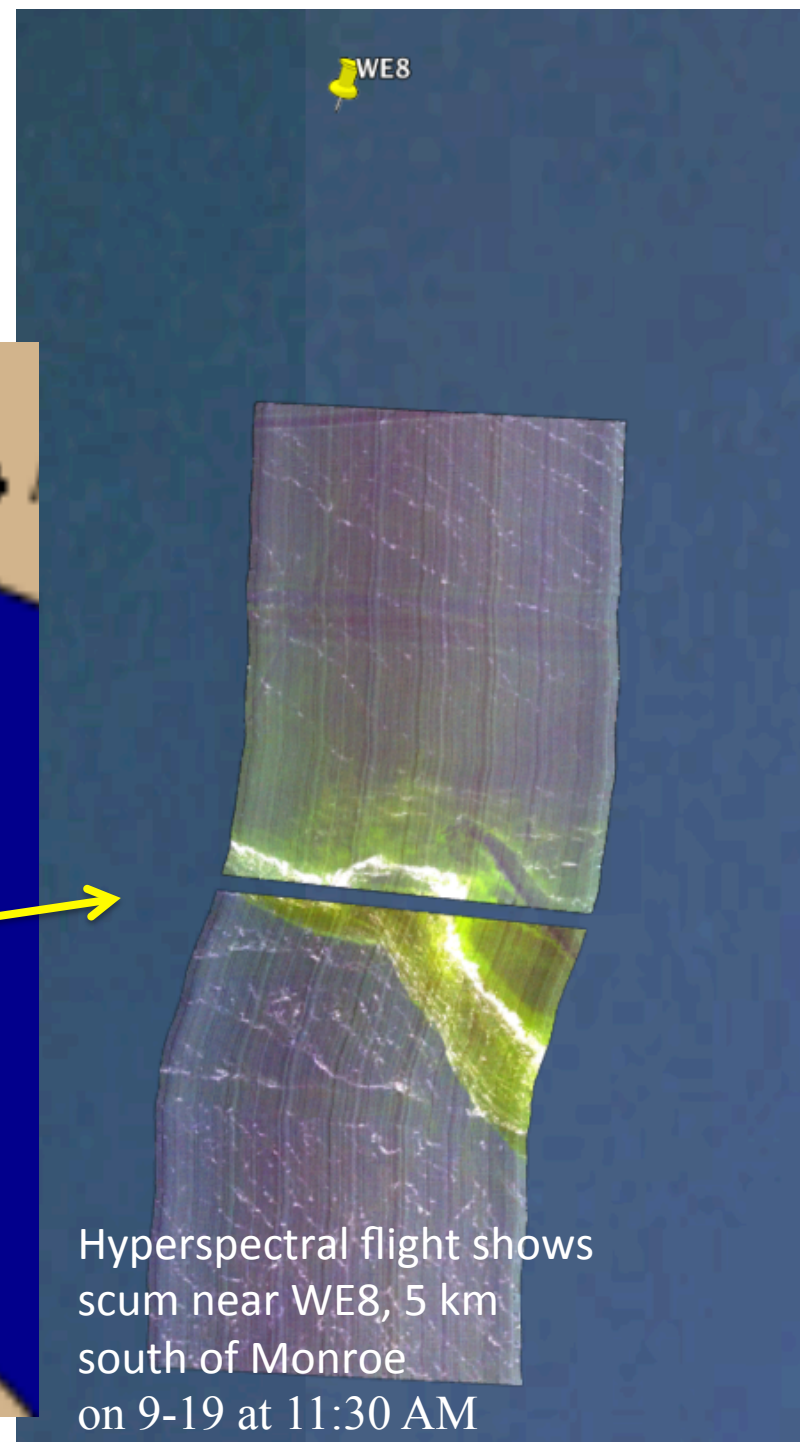
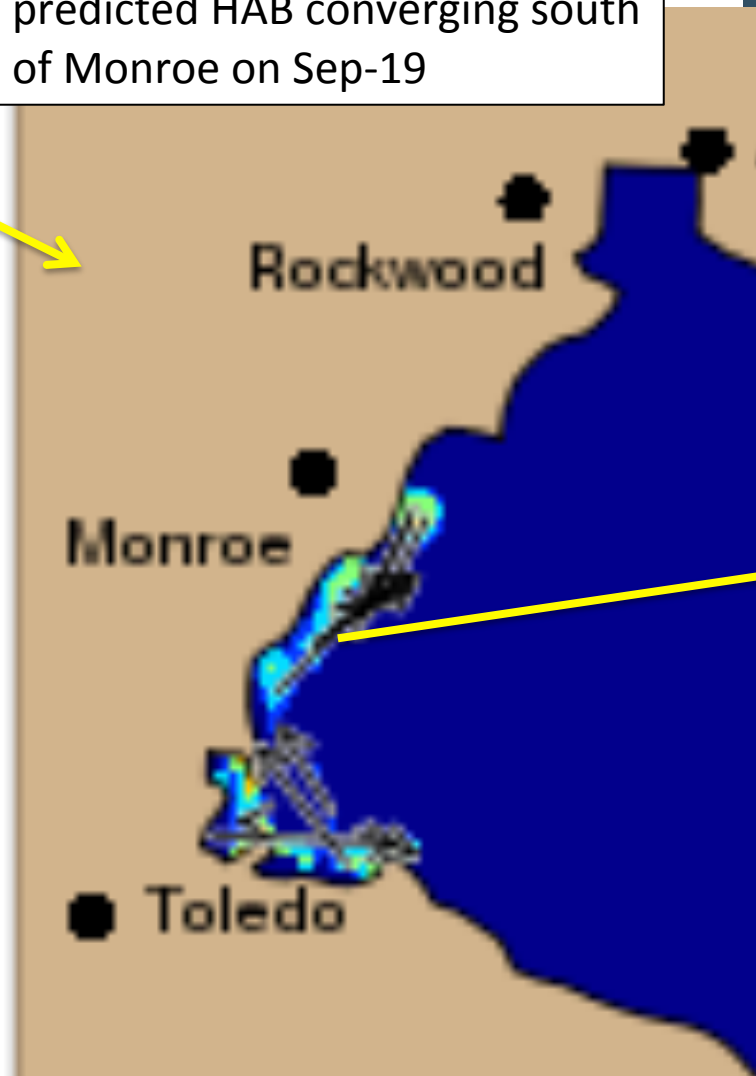
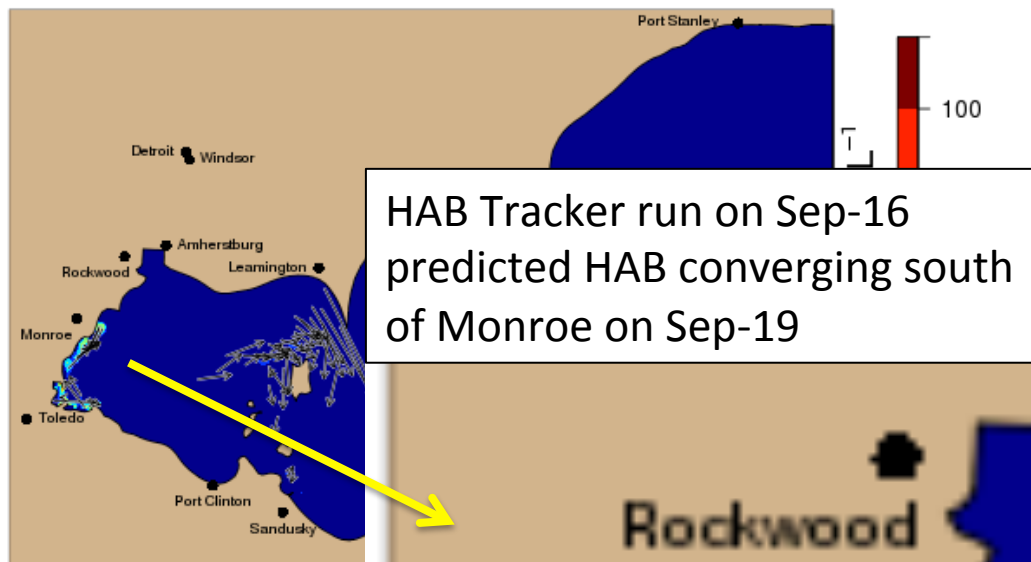
Vander Woude, AJ

17

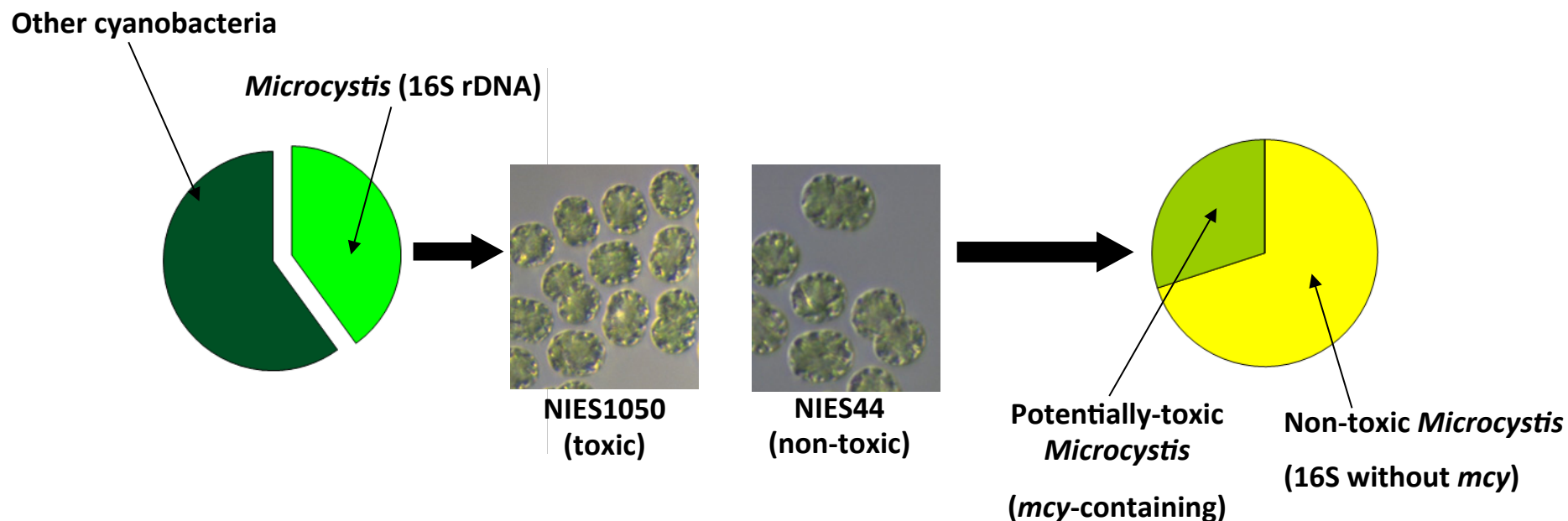


800 ft

2016-09-19 20:00 EDT



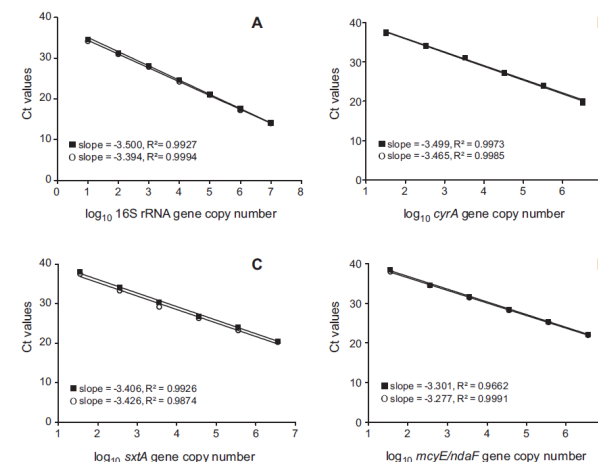
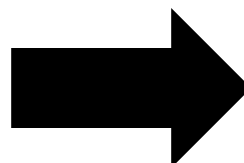
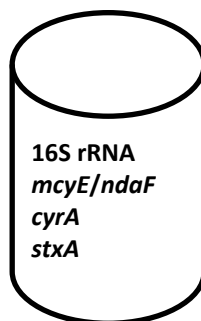
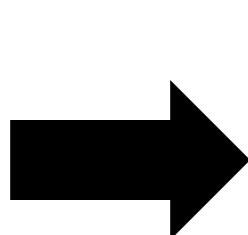
Using molecular tools to monitor CHABs:



% toxic *Microcystis* = proportion of *Microcystis* cells containing the genetics to produce microcystins ($mcyD$ or E / 16S x 100)

Determined by (1) qPCR and (2) metagenomics

Contact: Mark VanAsten, Mark.Vanasten@diagnostictechnology.com.au

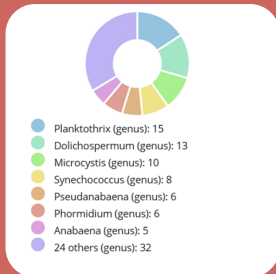


Where is this assay being used?

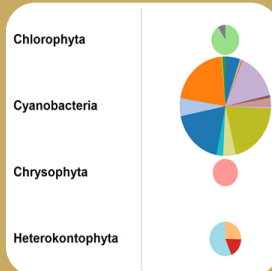
- USA
 - Part of Ohio EPA water testing algorithm rules
 - North East Ohio Regional Sewer District Laboratory (service offering)
 - Evaluations at Kansas (EPA Region 7) Californian Water Board, Fish & Game, Indiana Office of Water, US EPA Office R&D
- Australia
 - Sydney and Melbourne Water Utility, NSW Water
- Elsewhere
 - WLN, Netherlands
 - PUB Singapore

Rapid assessment of CHAB community composition using DNA Barcoding

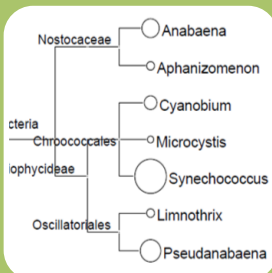
Work being conducted by Natalia Ivanova and Jerome Comte



Building 16S BOLD Reference Database for cyanobacteria and eukaryotic algae



Testing NGS protocols on mock communities generated from known cultures



Targeted NGS on field samples to simultaneously detect cyanobacteria and eukaryotic algae

What is omics?

process:	transcription → translation		
biomolecule:	DNA	mRNA	protein
cellular function:	encodes genes and stores hereditary information	an intermediate of genes and proteins	structural (e.g., cell wall) and catalytic (mediates biochemical reactions)
omics approach:	genomics	transcriptomics	proteomics
half-life:	days to years	minutes	hours to days
information provided:	the biological/geochemical potentials of an organism or community	which genes are actually being expressed	the identity and quantity of proteins present

Dick and Lam, *Elements Magazine*, in review



Article
pubs.acs.org/est

Nutrient-Controlled Niche Differentiation of Western Lake Erie Cyanobacterial Populations Revealed via Metatranscriptomic Surveys

Matthew J. Harkin,[†] Timothy W. Davis,[‡] Susan B. Watson,[§] and Christopher J. Gobler^{*,†}

The ISME Journal (2014), 1–13
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www.nature.com/ismej

ORIGINAL ARTICLE

Nutrients drive transcriptional changes that maintain metabolic homeostasis but alter genome architecture in *Microcystis*

Morgan M. Steffen¹, Stephen P. Dearth², Brian D. Dill^{3*}, Zhou Li^{3*}, Kristen M. Larsen¹, Shawn R. Campagna² and Steven W. Wilhelm¹

OPEN ACCESS Freely available online



Phylogenies of Microcystin-Producing Cyanobacteria in the Lower Laurentian Great Lakes Suggest Extensive Genetic Connectivity

Timothy W. Davis^{1,2*}, Susan B. Watson¹, Mark J. Rozmarynowicz², Jan J. H. Cilborowski², Robert Michael McKay², George S. Bullerjohn²

Genomic database will fuel research in the Great Lakes and beyond

Work being conducted by Greg Dick and Kevin Meyer, University of Michigan



Cyanohub BETA

Menu

- Cyanohub Home
- Query Form

Table of cultured *Microcystis* strains

Strain Name	Country	Toxic	Genome Size (Mb)	NCBI Tax. ID	No. Genes (documented)
PCC 7005	USA	No	4.9	267870	5081
PCC 7806	NETHERLANDS	Yes	5.3	267872	5169
PCC 9432	CANADA	No	5	1160280	4952
PCC 7941	CANADA	Yes	4.8	213618	4646
PCC 9443	CENTRAL AFRICAN REPUBLIC	Yes	5.1	1160281	5139
PCC 9701	FRANCE	No	4.7	721123	4673
PCC 9717	FRANCE	Yes	5.2	1160286	5234
PCC 9806	USA	No	4.2	1160282	4127
PCC 9807	SOUTH AFRICA	Yes	5.1	1160283	5104

Search Database

Basic Fields

Strain Name: Location:

- Searchable genomic database for CHAB genera including Great Lakes strains
- Publicly available resource
- Serves as a link between environmental and genomic data
- Critical to further understand the response of CHAB genera to environmental drivers

MBio Diagnostics Multiplexed Toxin Assay System

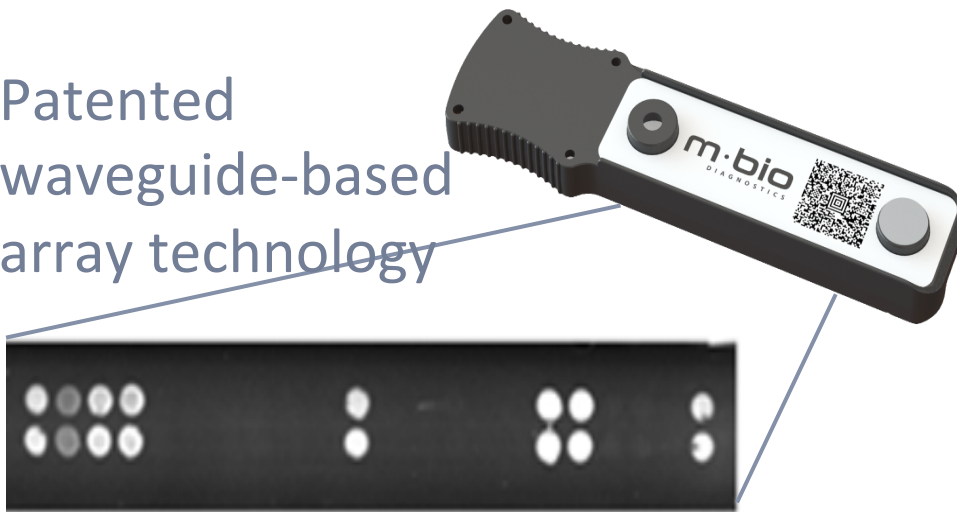
Contact: Mike Lochhead, mike.lochhead@mbiodx.com



- Cartridge-based multiplexed immunoassays for toxin detection
- Portable fluorescence reader
- Assay time < 15 minutes
- On-board reagents & waste



Patented
waveguide-based
array technology



Development of an inexpensive and user-friendly DNA preservation method

- Collaboration with George Bullerjahn, Steve Giglio, Susan Watson
- 12 strains were pipetted onto duplicate FTA cards and allowed to dry at room temperature and left at 24°C or 37°C for ~2 weeks
- DNA was then extracted and purified



Strain	Toxic?
<i>Anabaena variabilis</i> NIVA 19	+ mcy
<i>Synechococcus</i> sp. ARC 11	-
<i>Synechococcus</i> sp. CP1181	-
<i>Anabaena vigueri</i>	-
<i>Pseudanabaena</i> sp. LE011-01	-
<i>Planktothrix</i> sp. LE011-012	-
<i>Anabaena variabilis</i> NIVA 19	+ mcy
<i>Microcystis aeruginosa</i> 15A	unknown
<i>Anabaena planktonica</i>	-
<i>Anabaena</i> sp. A102	-
<i>Microcystis viridis</i> NIVA169/9	+ mcy
<i>Pseudanabaena limnetica</i> NIVA 111	unknown

Cyanobacterial DNA can be extracted and sequenced

Ambient (24°C)				Elevated (37°C)			
<u>Strain</u>	<u>Toxic?</u>	<u>cyano16S</u>	<u>mcyA</u>	<u>Strain</u>	<u>Toxic?</u>	<u>cyano16S</u>	<u>mcyA</u>
<i>Anabaena variabilis</i> NIVA 19	+ mcy	+	-	<i>Anabaena variabilis</i> NIVA 19	+ mcy	+	-
<i>Synechococcus</i> sp. ARC 11	-	+	-	<i>Synechococcus</i> sp. ARC 11	-	+	-
<i>Synechococcus</i> sp. CP1181	-	+	-	<i>Synechococcus</i> sp. CP1181	-	+	-
<i>Anabaena vigueri</i>	-	+	-	<i>Anabaena vigueri</i>	-	+	-
<i>Pseudanabaena</i> sp. LE011-01	-	-	-	<i>Pseudanabaena</i> sp. LE011-01	-	-	-
<i>Planktothrix</i> sp. LE011-012	-	-	-	<i>Planktothrix</i> sp. LE011-012	-	-	-
<i>Anabaena variabilis</i> NIVA 19	+ mcy	+	+	<i>Anabaena variabilis</i> NIVA 19	+ mcy	+	+
<i>Microcystis aeruginosa</i> 15A	unknown	+	-	<i>Microcystis aeruginosa</i> 15A	unknown	+	-
<i>Anabaena planktonica</i>	-	+	-	<i>Anabaena planktonica</i>	-	+	-
<i>Anabaena</i> sp. A102	-	+	-	<i>Anabaena</i> sp. A102	-	+	-
<i>Microcystis viridis</i> NIVA169/9	+ mcy	+	+	<i>Microcystis viridis</i> NIVA169/9	+ mcy	+	+
<i>Pseudanabaena limnetica</i> NIVA 111	unknown	+	+	<i>Pseudanabaena limnetica</i> NIVA 111	unknown	+	+

- DNA was extracted from 83% of samples with 100% amplification of the cyano16S gene in all samples yielding DNA and 2 of the 3 of known toxic strains
- PCR products from *Synechococcus* sp. ARC 11 was sequenced for verification
- Cards are cheap, easy to use, do not need specialized storage, and inexpensive to send
- FTA cards are a valuable tool for water managers, park rangers and citizen scientists