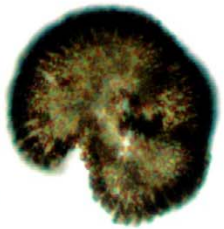
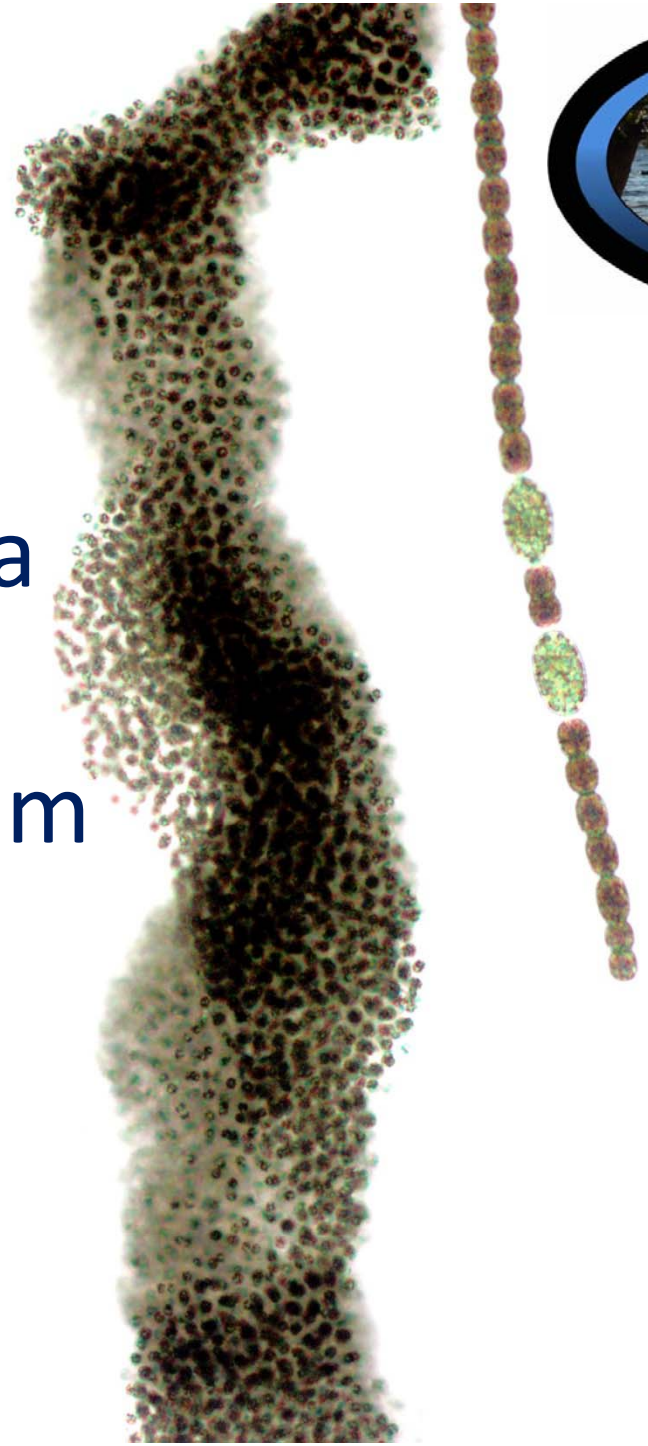




Center for
UNH
CFB
Freshwater Biology

CFB.UNH.EDU

Development of a Cyanobacteria Monitoring Program

Amanda L. Murby

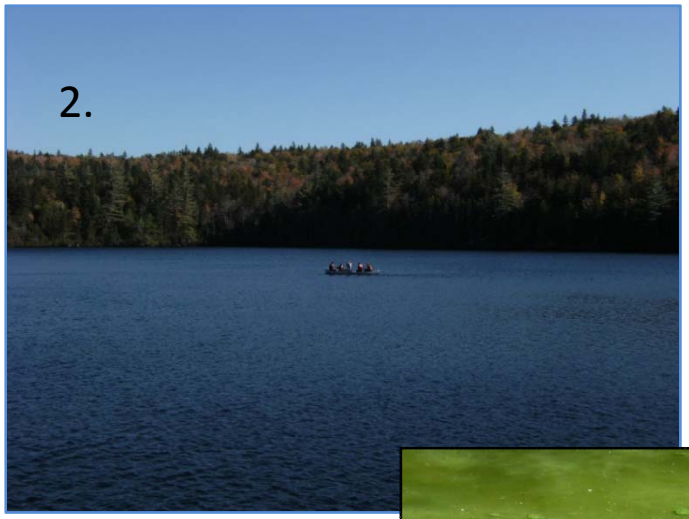
April 14, 2010

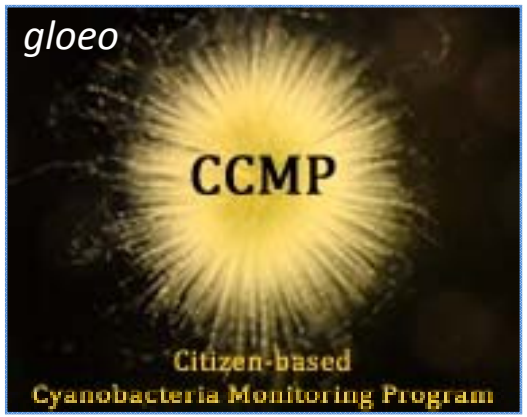
NEIWPCC
New England Interstate Water Pollution Control Commission

50 µm

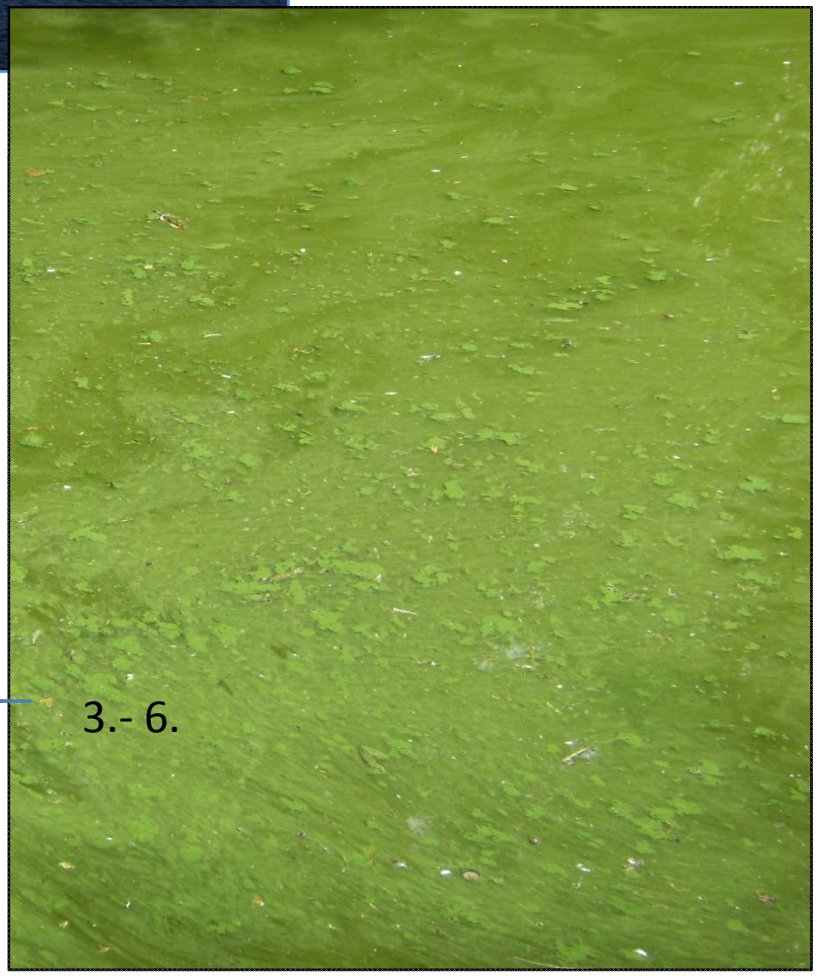
Questions?

“ who does testing for cyanotoxins ?”
“ which cyanotoxins can be detected ?”
“ why are there no regulations on cyanotoxins in US freshwaters ?”
“ how can our lake association monitor for cyanotoxins ?”
“ what can we do to control blooms?”
“ where can I test my water for cyanotoxins ?”





1. Drinking Water
2. Lake Monitoring
3. Blooms
4. Plant Tissues
5. Animal Tissues
6. Aerosols



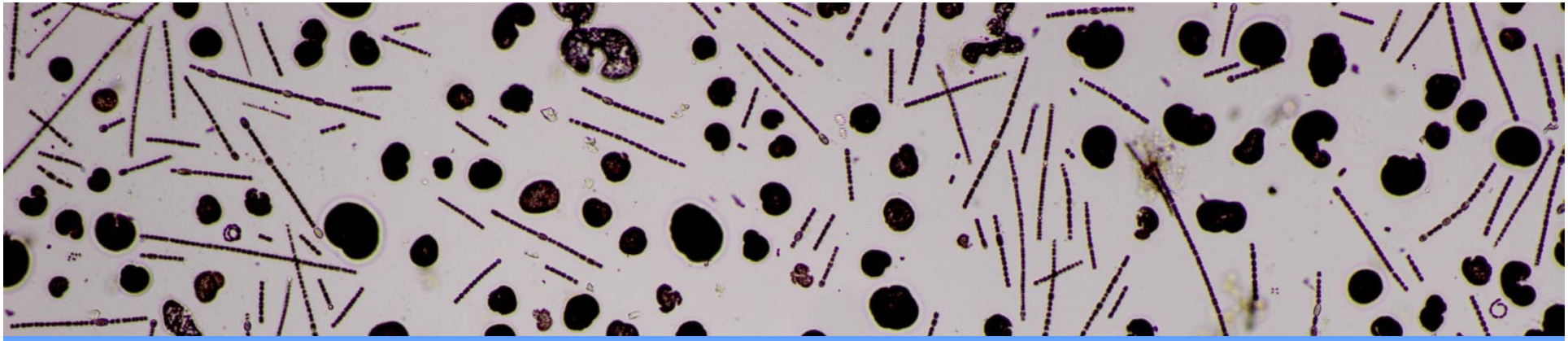
Limits for contact with microcystins

Table 1. Toxicity of cyanobacterial toxins.

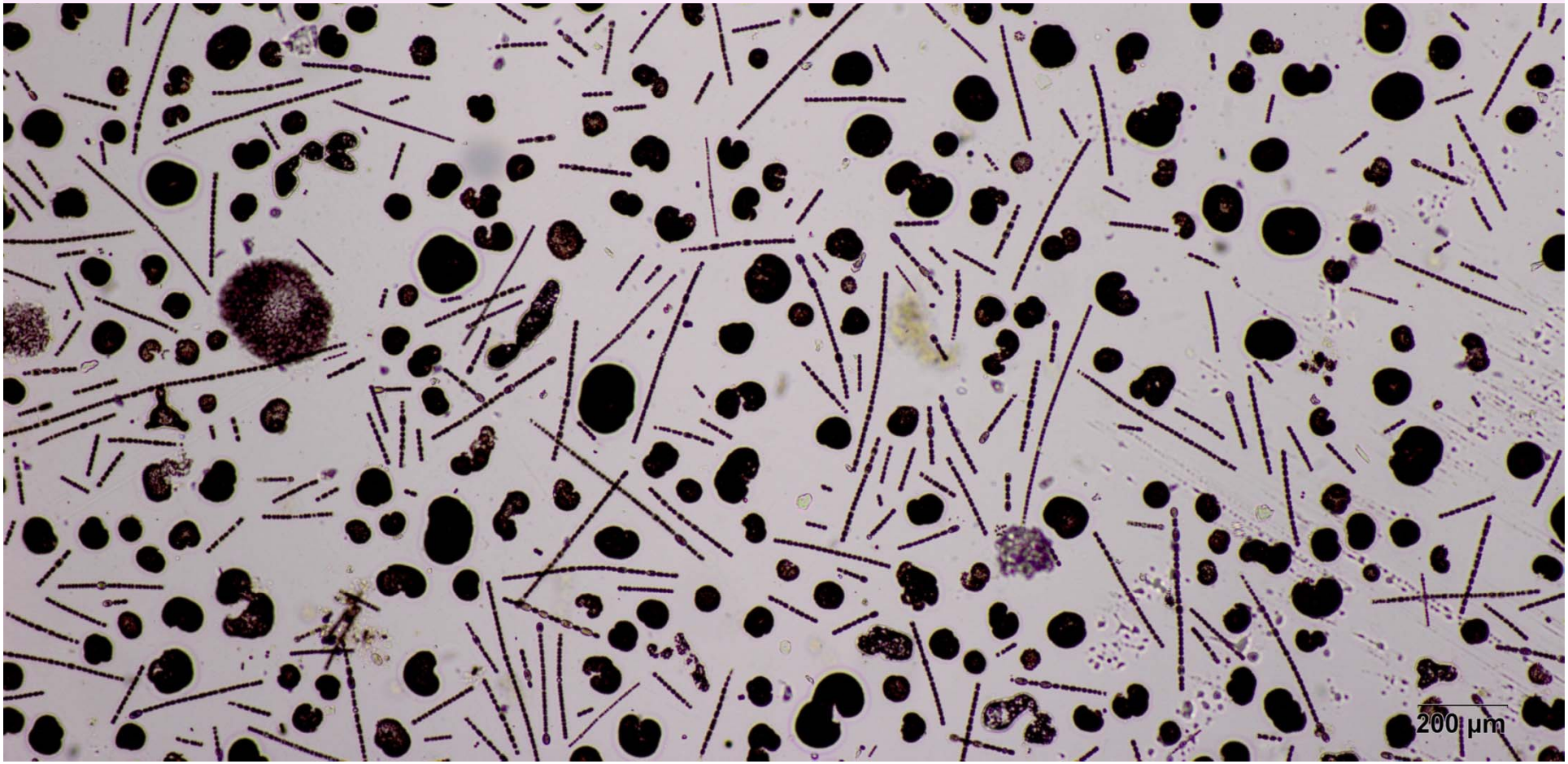
Toxin	LD_{50} (μg/kg, ip, mouse)	Organism	Reference
Microcystin-LR	50	*M. aeruginosa, Aph. flos-aquae, M. viridis*	(31,125)
Microcystin-LA	50	*M. aeruginosa, M. viridis*	(138)
Microcystin-YR	70	*M. aeruginosa, M. viridis*	(31)
Microcystin-RR	600	*M. aeruginosa, Anabaena sp., M. viridis*	(139–141)
[*D*-Asp^3]microcystin-LR	50–300	*M. aeruginosa, Aph. flos-aquae, M. viridis, O. agardhii*	(142,143)
[*D*-Asp^3]microcystin-RR	250	*O. agardhii, M. aeruginosa, Anabaena sp.*	(19,139)
[*Dha*^{7}]microcystin-LR	250	*M. aeruginosa, Anabaena sp., O. agardhii*	(139,144)
[(6Z)-Adda]microcystin-LR	> 1200	*M. viridis*	(143)
[(6Z)-Adda]microcystin-RR	> 1200	*M. viridis*	(143)
Nodularin	50	*N. spumigena*	(145)
[*D*-Asp^1]nodularin	75	*N. spumigena*	(146)
[(6Z)-Adda^5]nodularin	> 2000	*N. spumigena*	(146)
Anatoxin-a	200–250	*Aph. flos-aquae, Anabaena spp., Oscillatoria sp., Aphanizomenon sp., Cylindrospermum sp.*	(145,147)
Anatoxin-a(s)	20	*Aph. flos-aquae*	(148)
Saxitoxin	10	*Aph. flos-aquae, A. circinalis, Cylindrospermopsis raciborskii, Lyngbya wollei*	(42,149)
Cylindrospermopsin	2000	*C. raciborskii, Umezakia natans, Aph. ovalisporum*	(150)

1. Drinking Water: 1 μg MC L^{-1}
2. Lake Monitoring: 20 μg MC L^{-1}
3. Blooms: 20 μg MC L^{-1}
4. Plant & Animal Tissues: (13 ng MC g^{-1} dwt fish tissue).
5. Aerosols: no guidelines

- Guidelines are based on WHO and MADPH thresholds http://www.mass.gov/Eeohhs2/docs/dph/environmental/exposure/protocol_cyanobacteria.pdf
- MC i.p. LD_{50} range from 25-150 μg MC kg^{-1} (mice)
- Neurotoxins are more potent… anatoxins are 10 μg kg^{-1} and Saxitoxins are 20 μg kg^{-1}
- Oral consumption is even more toxic for humans at 5- 10 μg MC kg^{-1}



Identification is important for determining potential cyanotoxicity

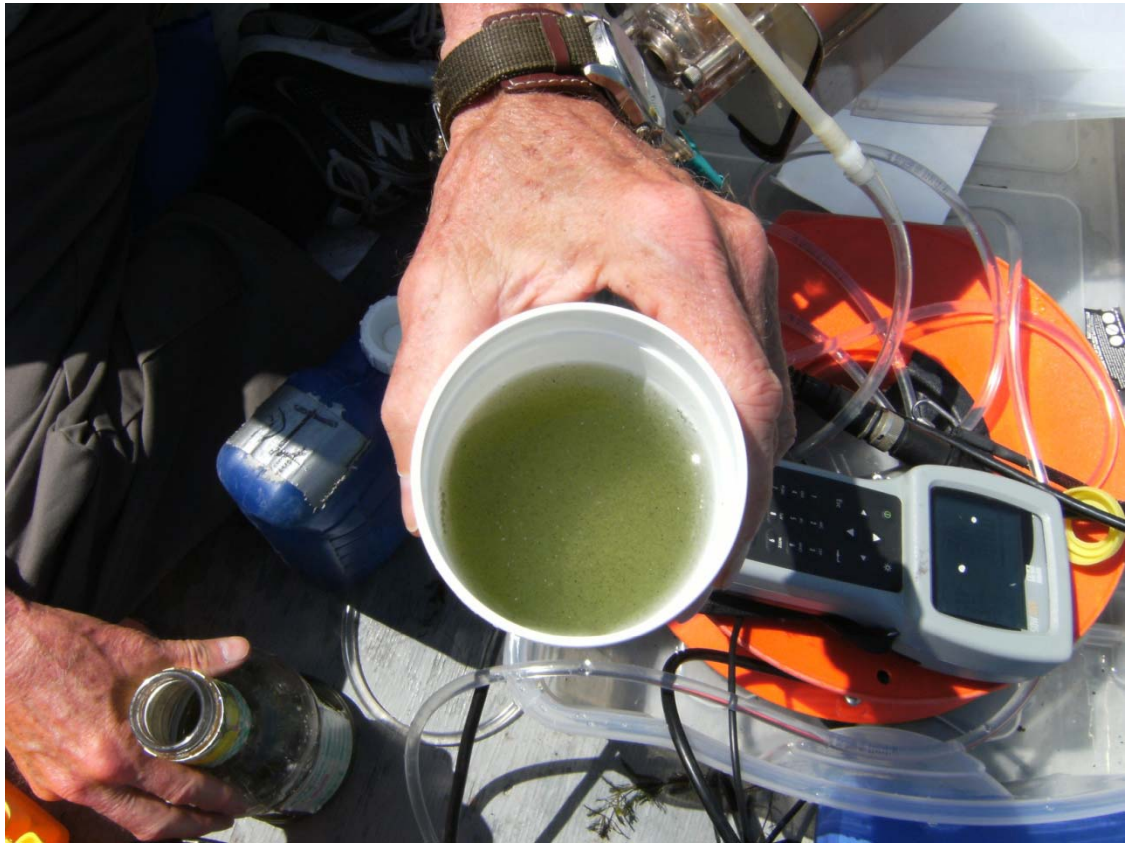


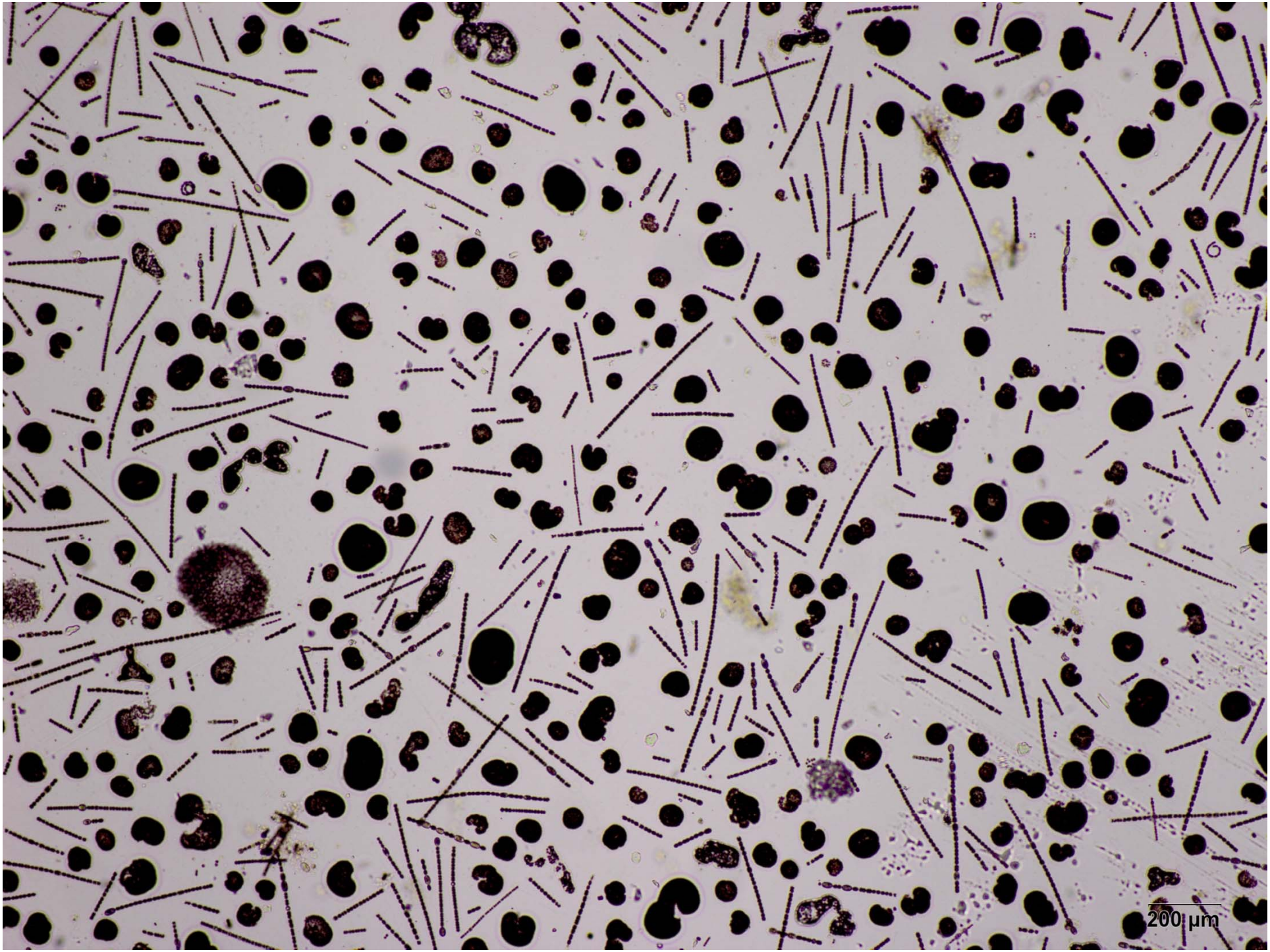
World Health Organization recommendation

- “In regions critical with respect to current or future use of ponds as drinking-water source, further studies are necessary for a more complete assessment of potential exposure risks. These should follow monthly or fortnightly sampling regimes. For this purpose, microscopic counting techniques should be applied for identification of species and for quantifying cyanobacterial cell density”.

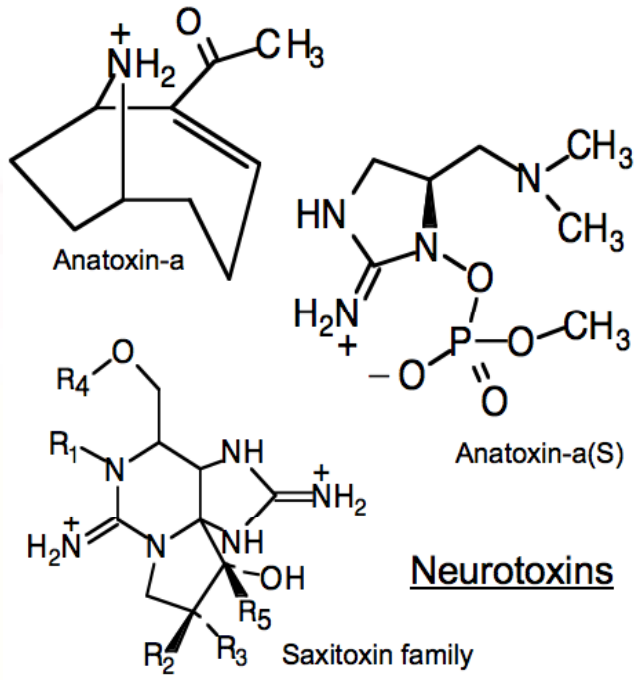
- Welker, Chorus, Fastner 2004

- Extreme blooms can be lethal for a 10 kg child if even just 100 ml (less than half of a cup) were consumed.




200 µm

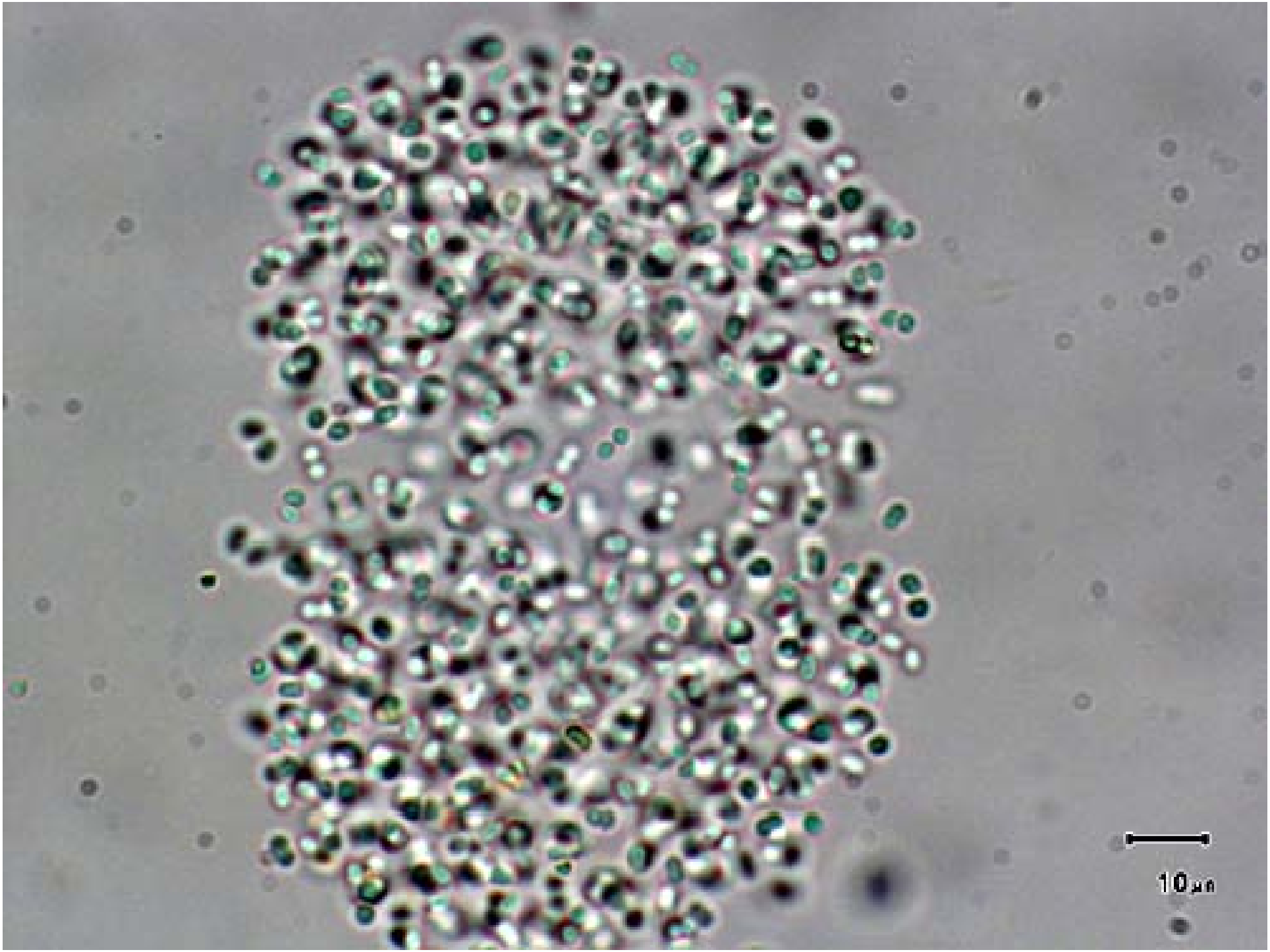
Aphanizomenon

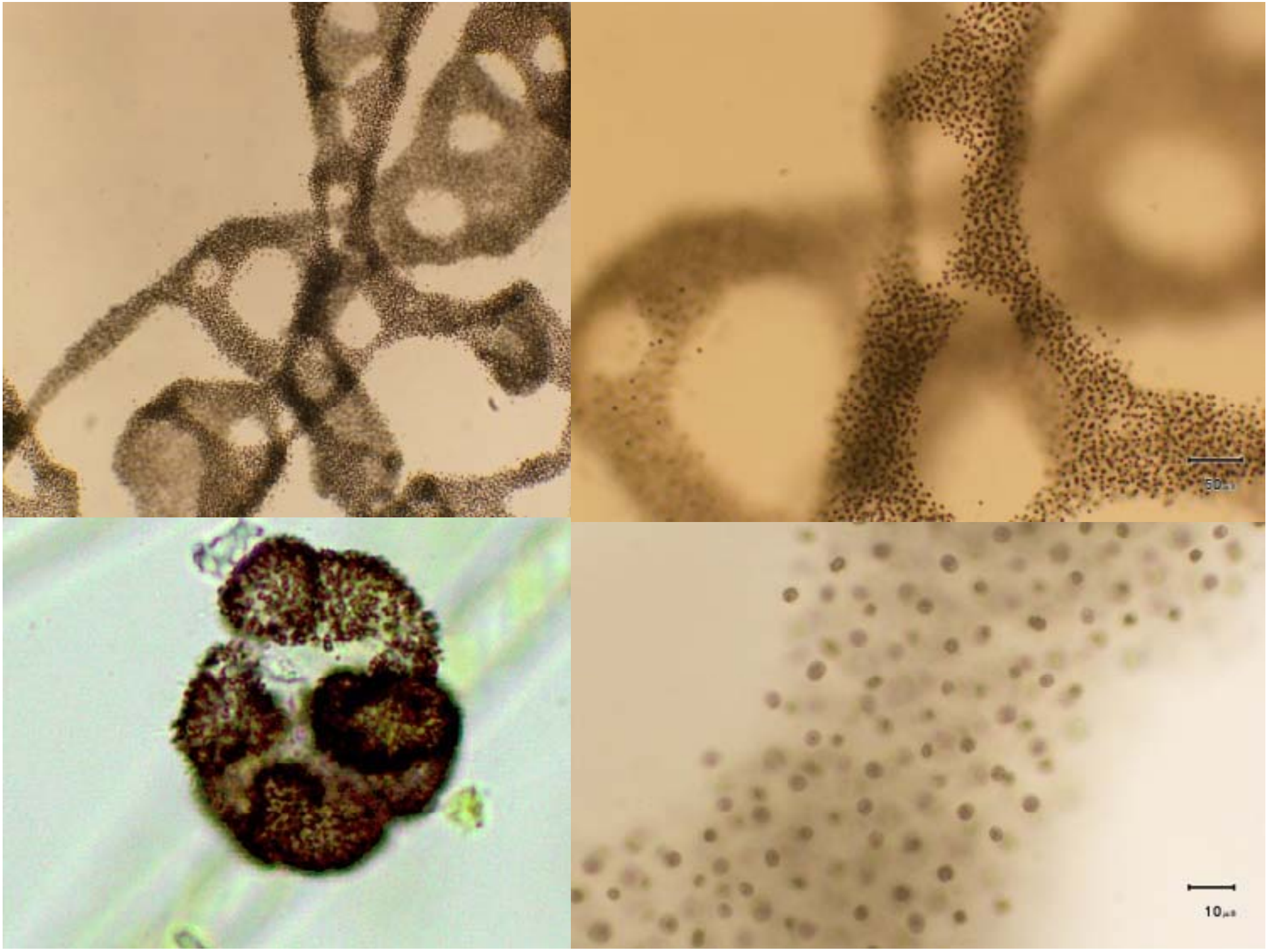


Neurotoxins


UNH CFB


UNH CFB

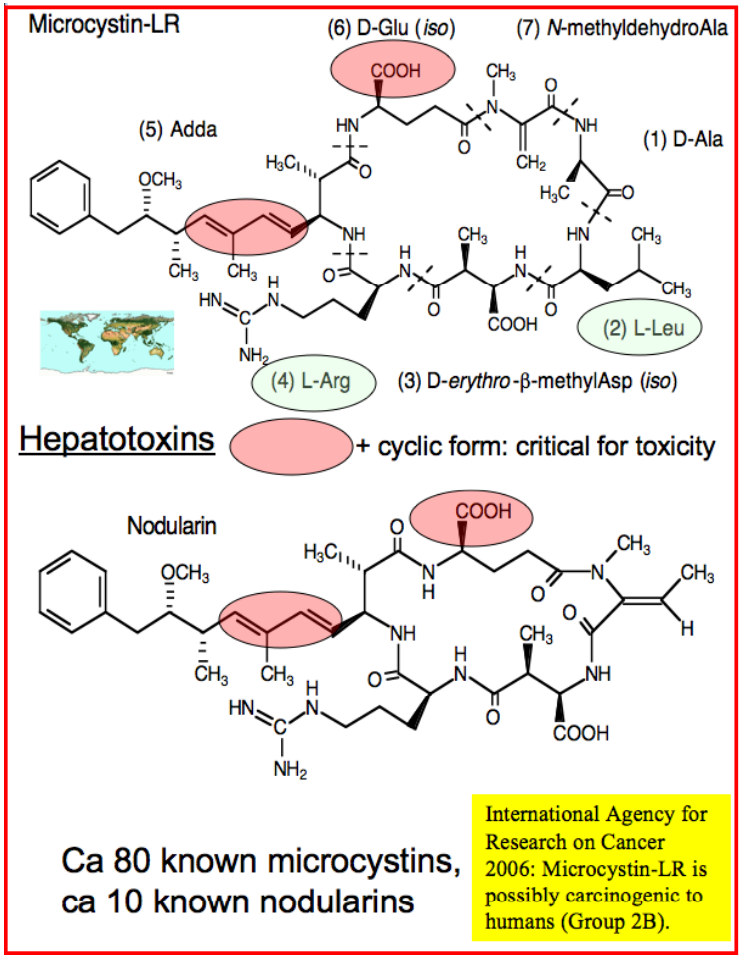

10 µm

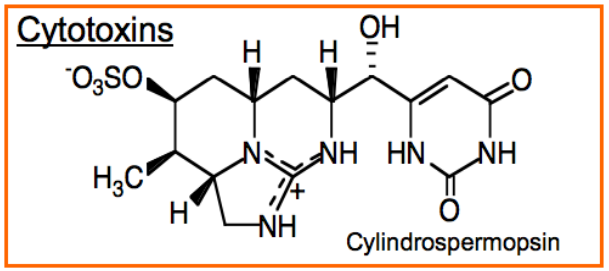

50µm
10µm

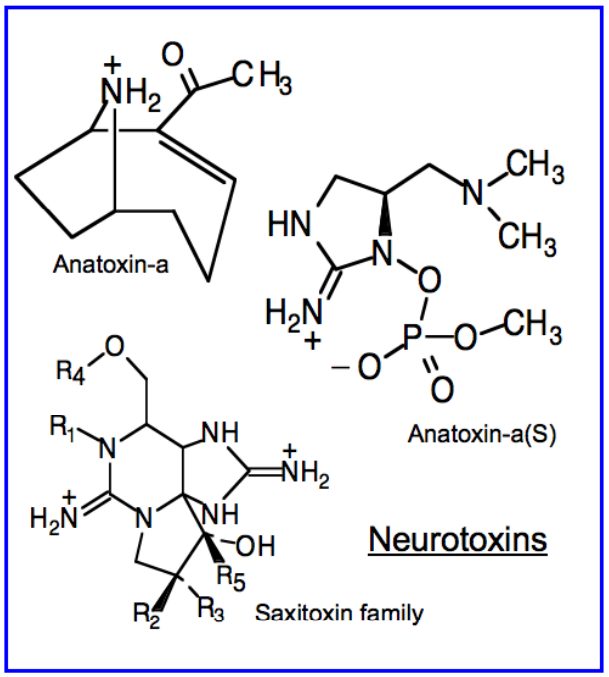
It is important to note that microcystins, even at low levels, provide a warning that toxigenic cyanobacteria are present. Furthermore, the level of a single toxin (such as microcystins), is generally an underestimate of the total risk to human health since most lake-cyanobacteria are capable of producing more than one type of toxin.

Most common and widespread of the cyanotoxins…

Microcystins as a proxy for other cyanotoxins?



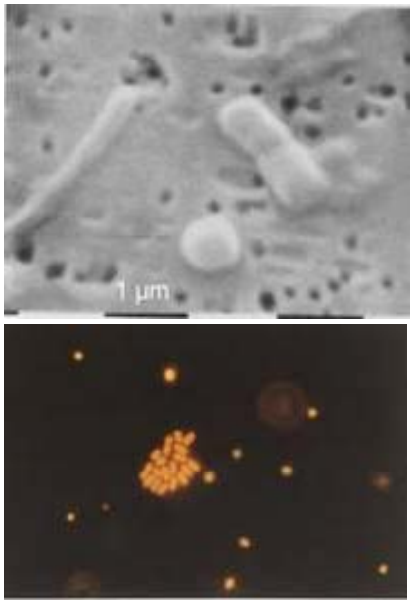


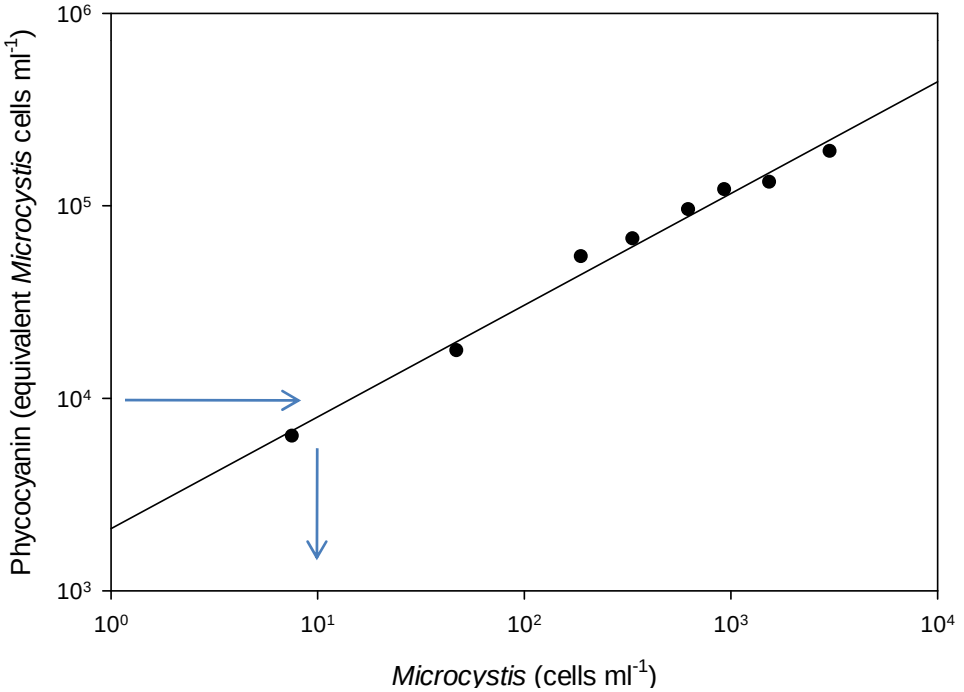


http://users.abo.fi/jmeriluo/

The use of phycocyanin for determining cells of cyanobacteria *in situ*

picocyanobacteria

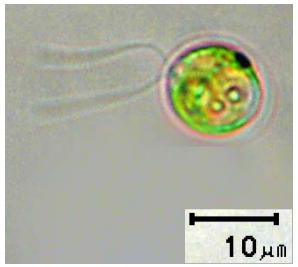




Phycocyanin (PC) Fluorescence

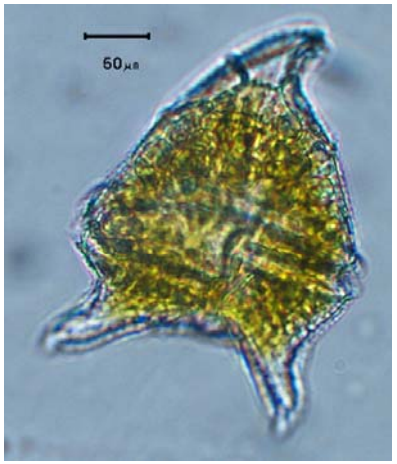
Pros:

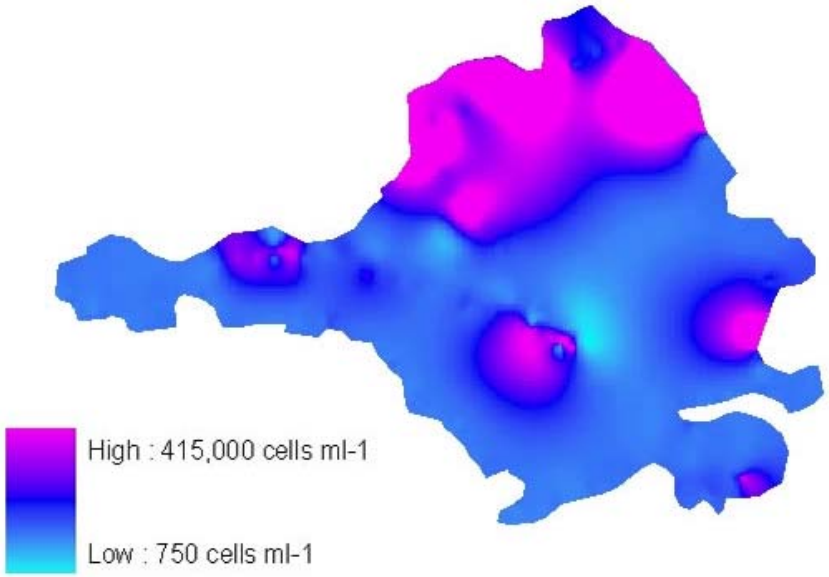
- Quick assessment and a good predictor of cyanobacteria
- Measures all sizes of cyano-cells (even the "picos", capable of producing MC and/or BMAA)

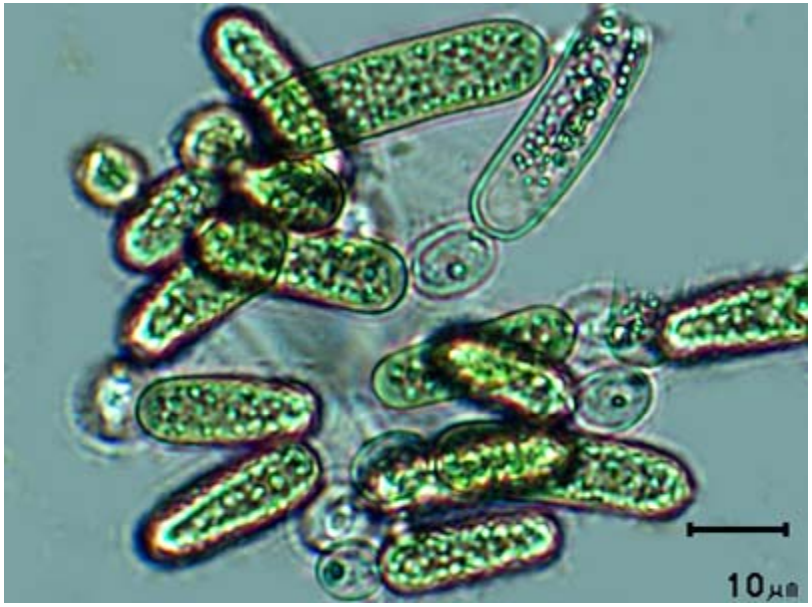


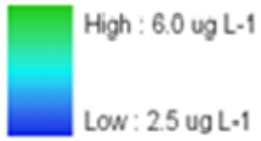
Cons:

- Temperature sensitive (of the response from phytoplankton)
- *Chlamydomonas* and some dinoflagellates
- Still necessary to ID plankton for reference to abundance and toxin production.

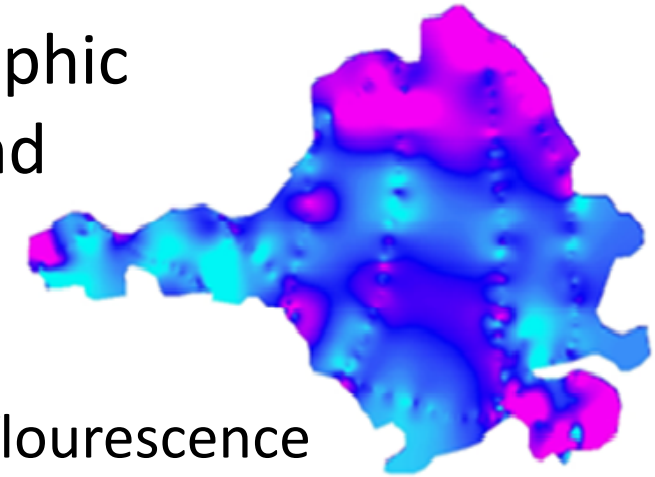






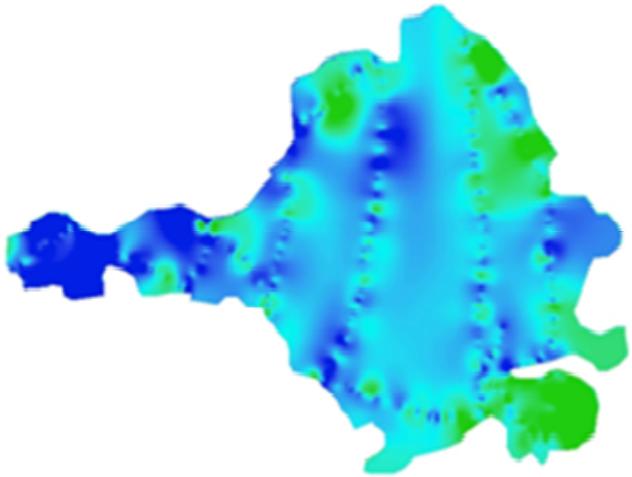


Meso-eutrophic
Willand Pond

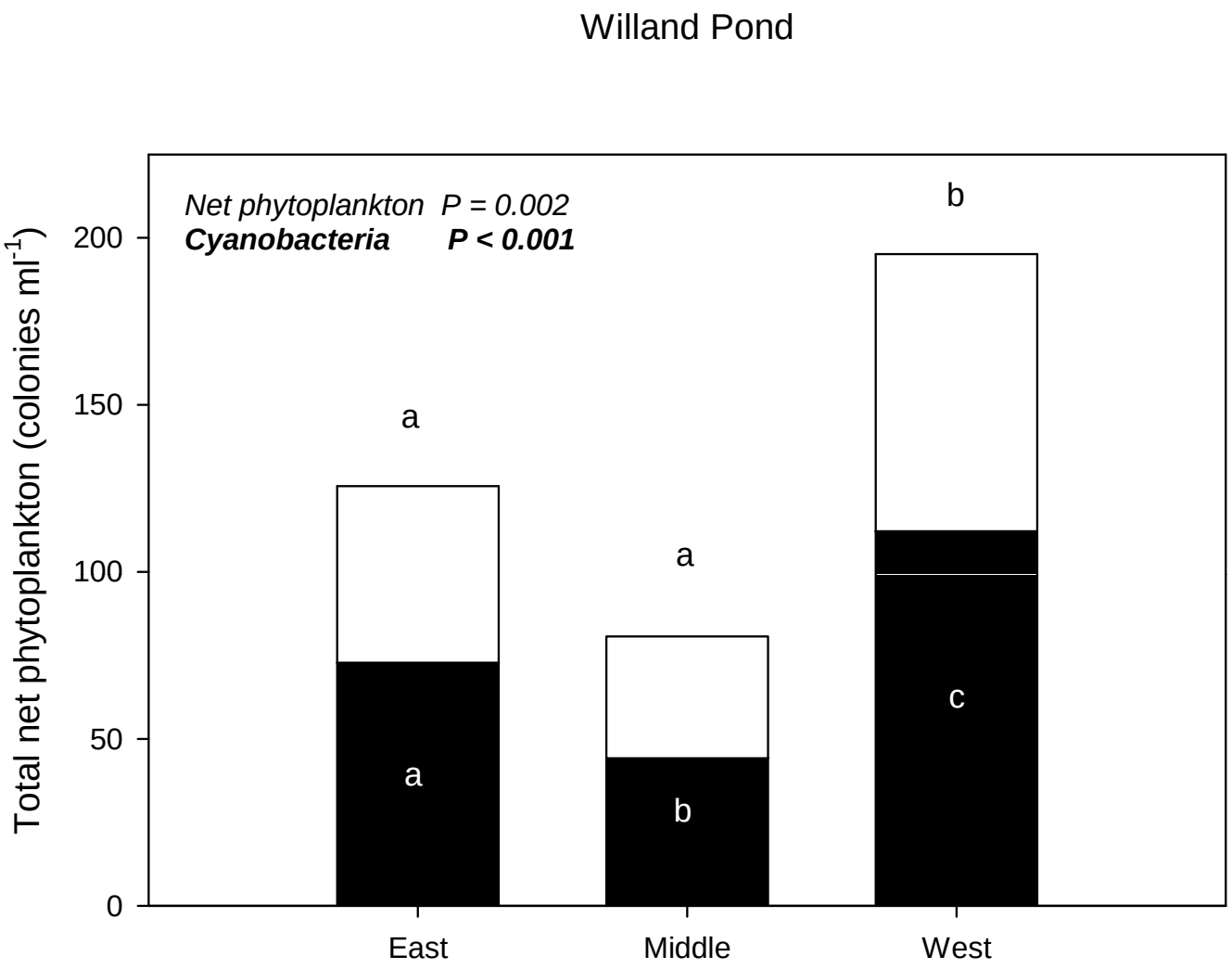


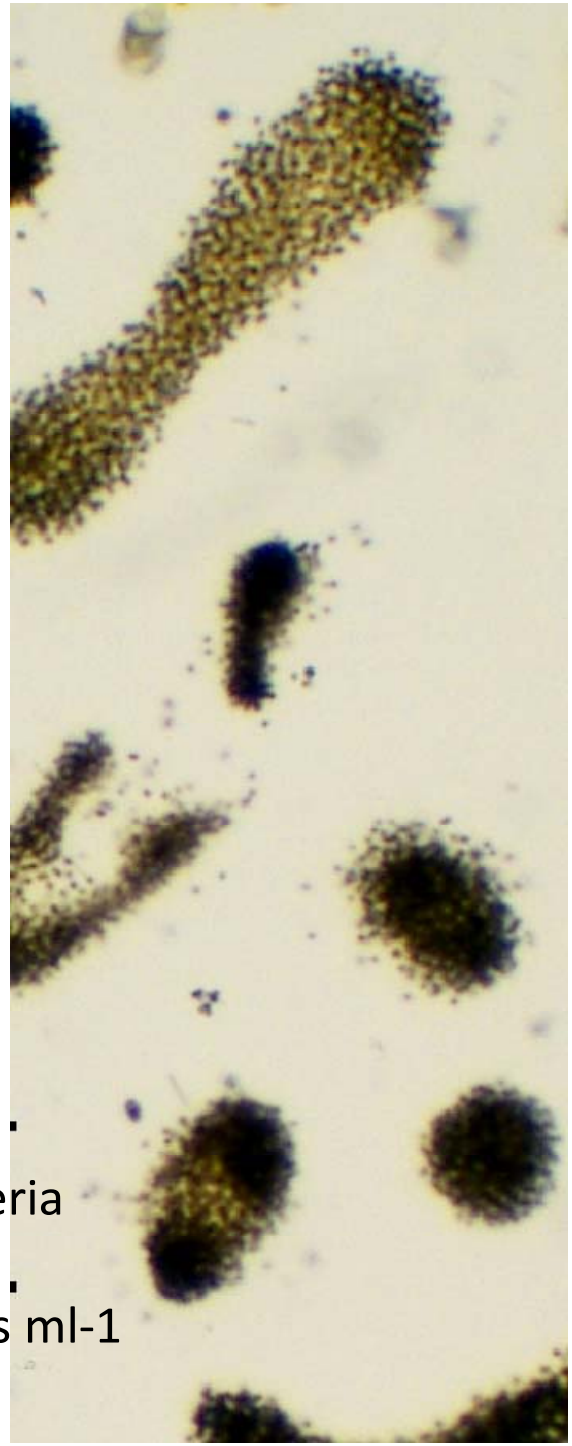
Phycocyanin flourescence

Cyanobacteria



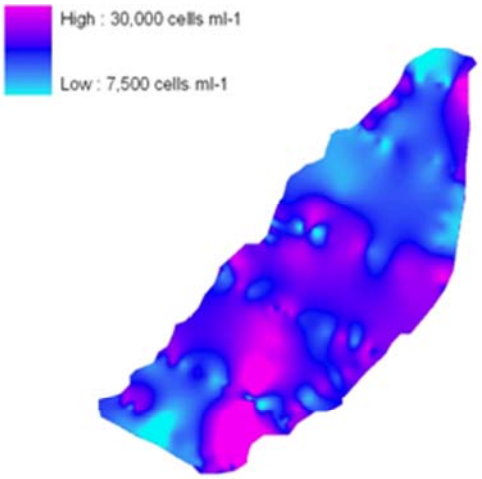
Chlorophyll



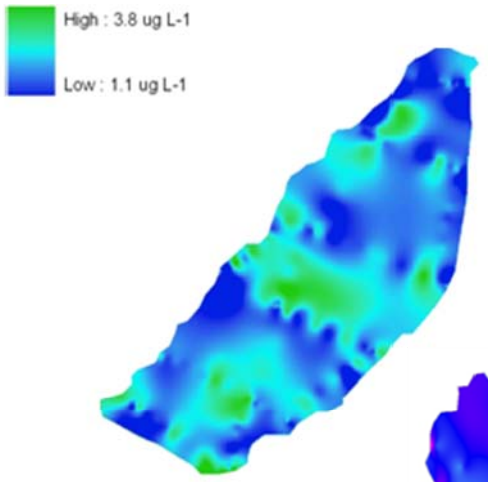


Variation in abundance of net phytoplankton and cyanobacteria

Each colony of *Microcystis* may have 100's to 1000's of cells ml-1



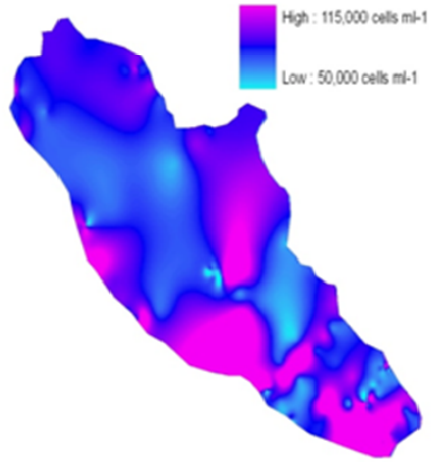
Cyanobacteria



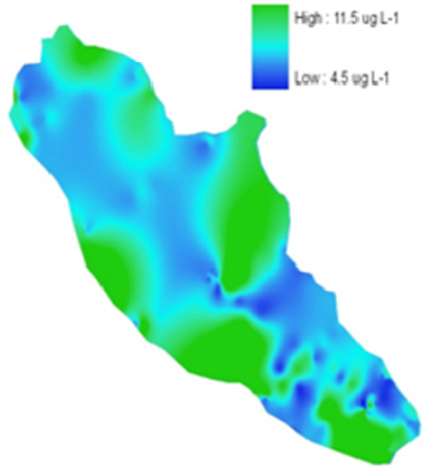
Chlorophyll

Eutrophic
Barbadoes Pond

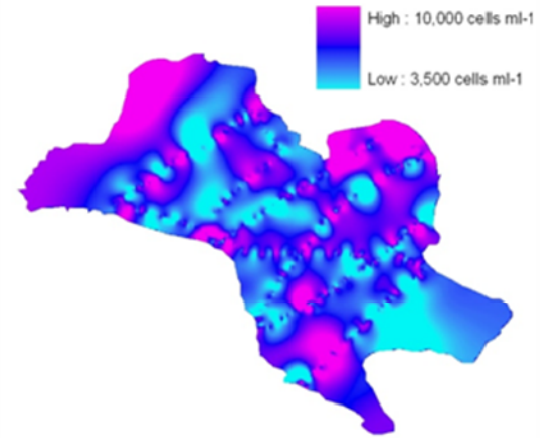
Mesotrophic
Silver Lake



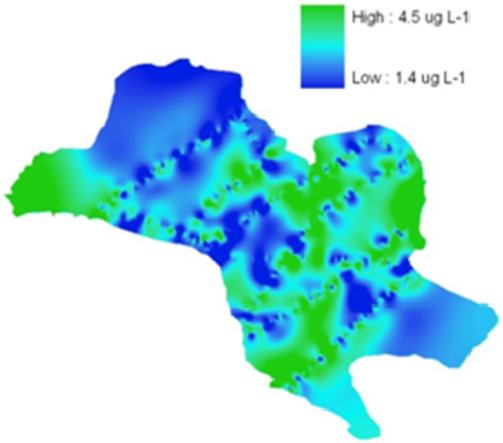
Cyanobacteria



Chlorophyll

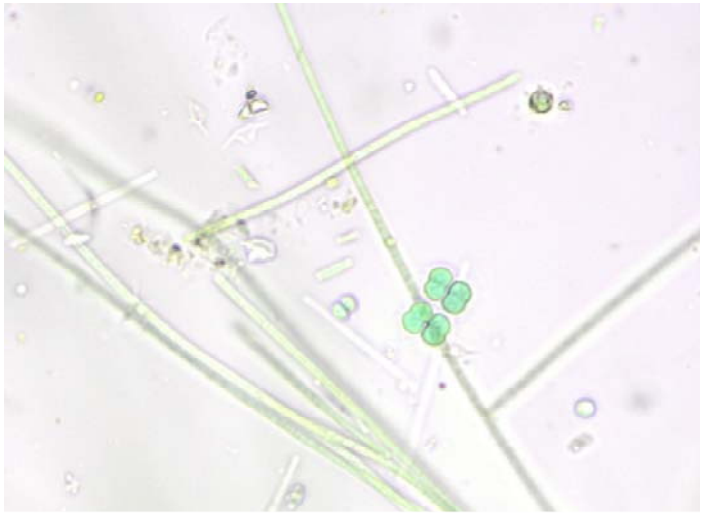


Cyanobacteria

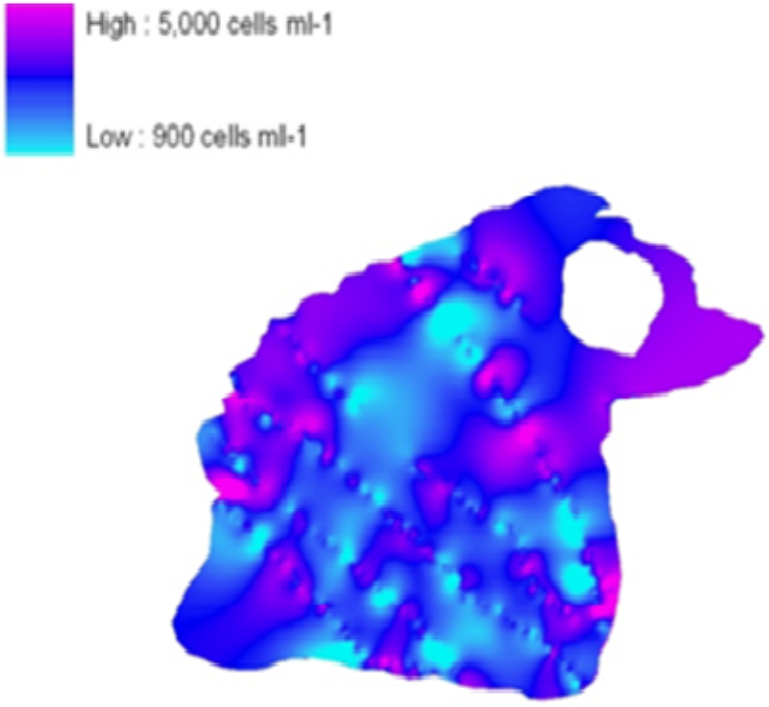


Chlorophyll

Meso-oligotrophic
Mirror Lake

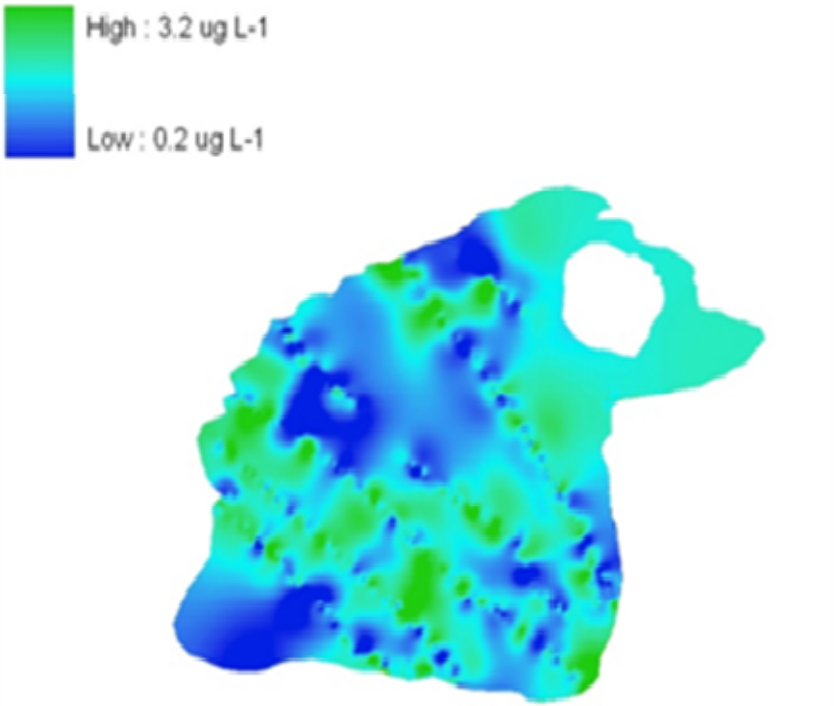


Ultra-oligotrophic Sawyer Pond

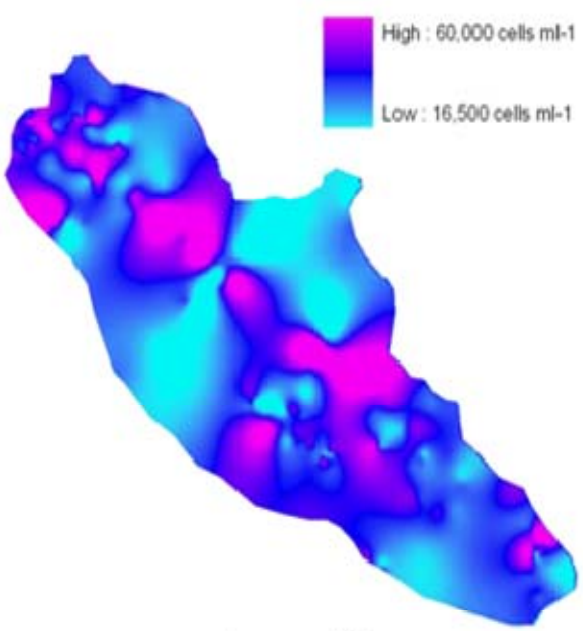


Phycocyanin flourescence

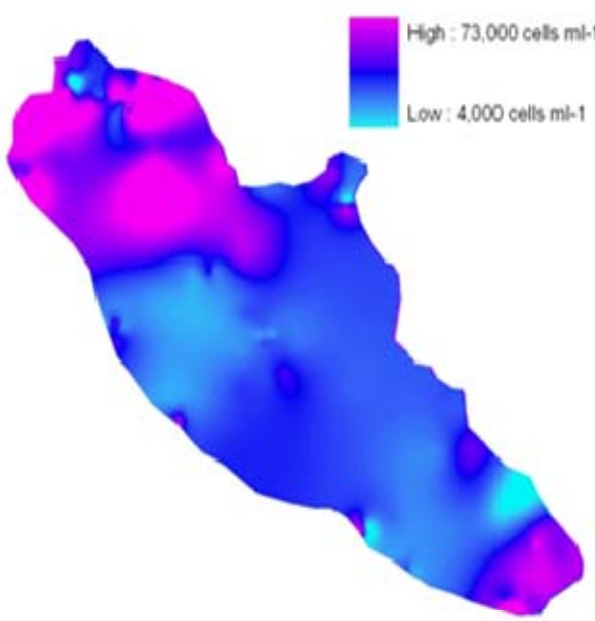
Cyanobacteria



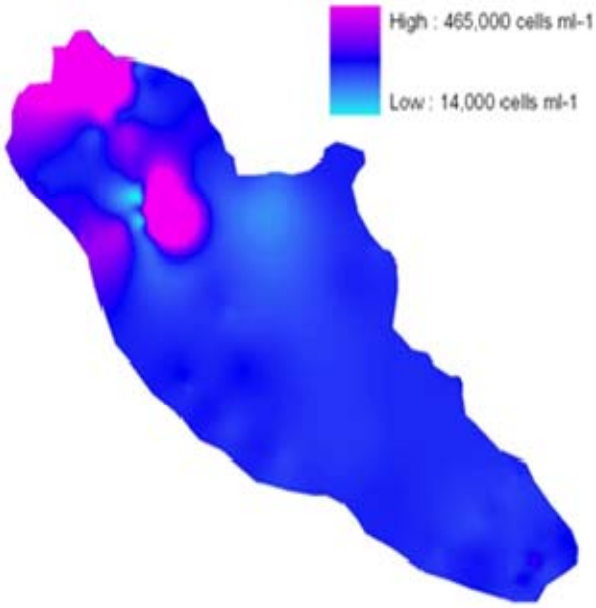
Chlorophyll



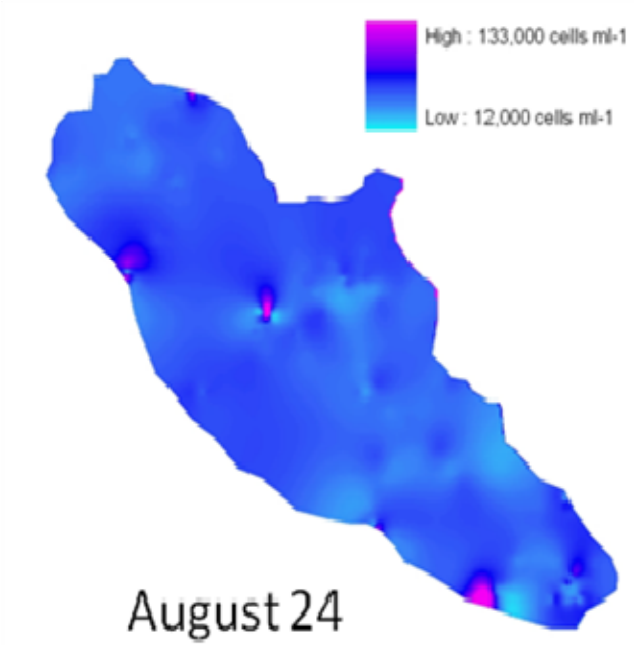
June 21



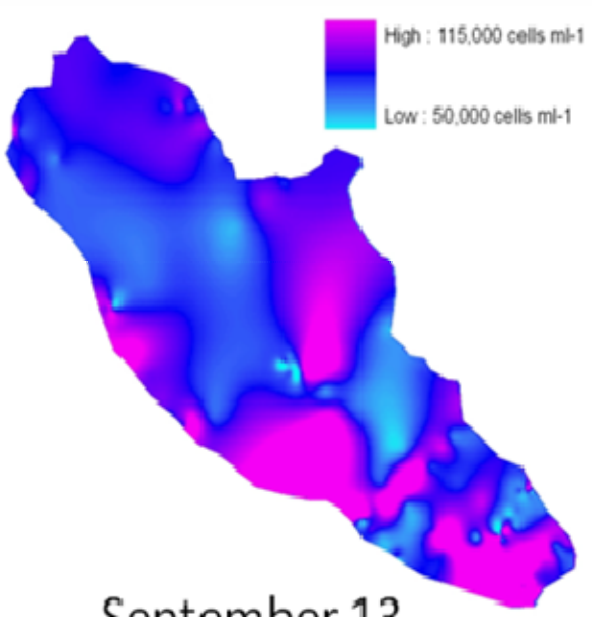
July 10



July 21



August 24

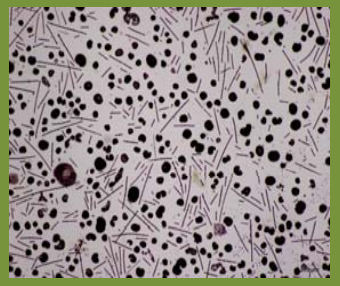


September 13

1. Synoptic lake sampling
2. Tracking bloom events
3. Testing of drinking water

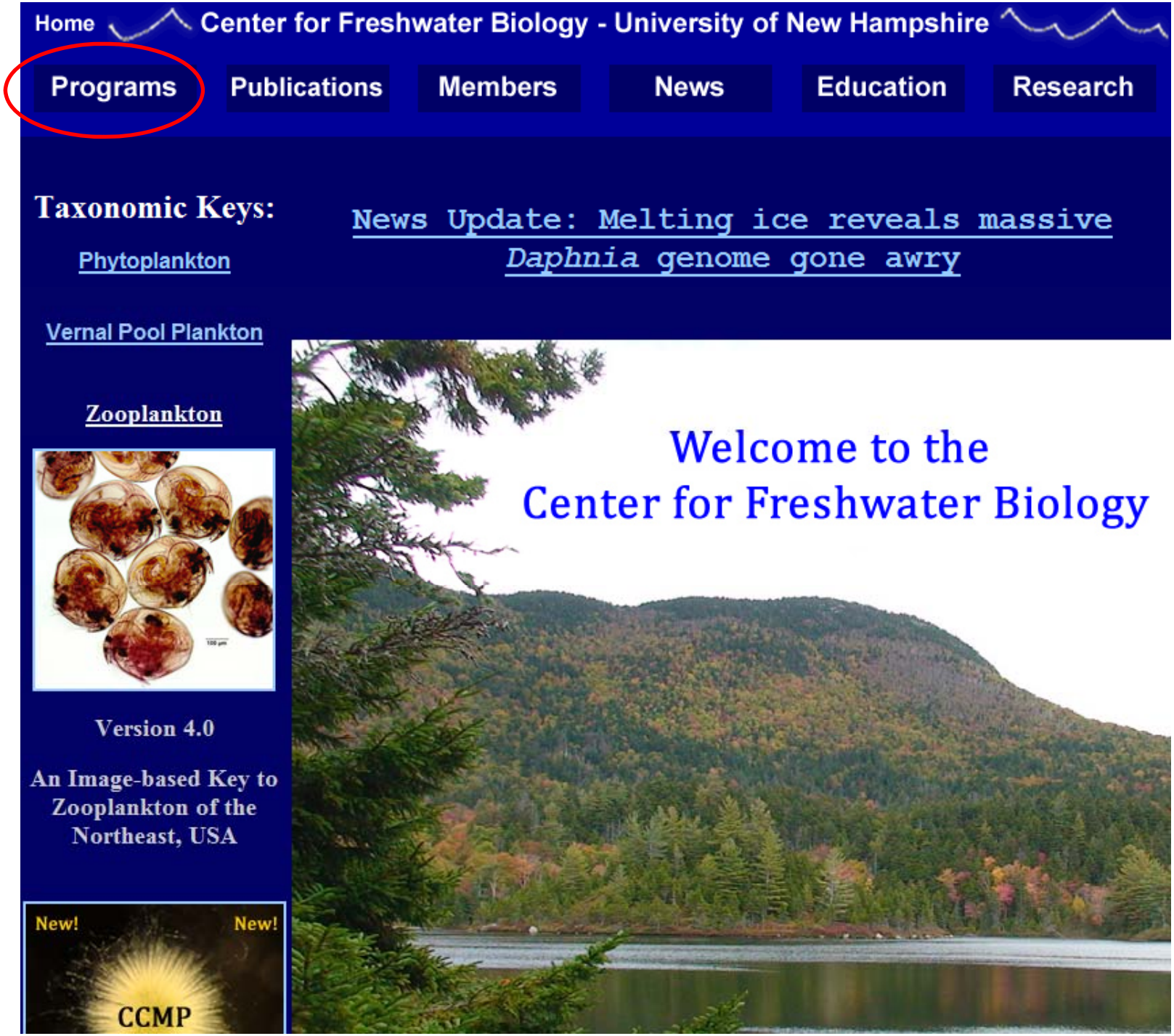
1. **Routine lake sampling** is important for tracking trends of cyanobacteria and microcystins in various types of water bodies.

2. **Visual surface "blooms" of cyanobacteria** are important to sample since they often have higher concentrations of cyanobacteria than are present in the open water due to an accumulation effect. If possible, take a picture and submit it electronically in addition to the water sample as it may help identify the dimensions of the bloom.



3. **Drinking water** was tested at different stages of processing, highlighting comparisons of microcystins from both, raw and finished water.

Protocols


Home
Center for Freshwater Biology - University of New Hampshire
Programs
Publications
Members
News
Education
Research
Taxonomic Keys:
Phytoplankton
Vernal Pool Plankton
Zooplankton
Version 4.0
An Image-based Key to Zooplankton of the Northeast, USA
New!
New!
CCMP
News Update: Melting ice reveals massive Daphnia genome gone awry
Welcome to the
Center for Freshwater Biology

Programs

Citizen-based Cyanobacteria Monitoring Program

Sampling protocols and services provided by the Center for Freshwater Biology (UNHCFB CCMP):

Cyanobacteria Lake Monitoring

Cyanotoxins in Drinking Water

The Center for Freshwater Biology at the University of New Hampshire has initiated a Citizen-based Cyanobacteria Monitoring Program (CCMP), providing assistance to Lake Associations and Drinking Water Facilities in the tracking of cyanobacteria and microcystins in various bodies of water. Protocols and services can be accessed by clicking the links at the left of the panels.

Please contact Amanda Murby for questions or assistance.

CCMP General Procedure for the Detection of Cyanobacteria and Microcystins:

- Frozen water samples were sent to the UNH, CFB.

- Phycocyanin fluorescence: **Cyanobacteria**

Samples were thawed and mixed thoroughly prior to measurements of relative concentrations of cyanobacteria *via* phycocyanin fluorescence (calibrated as equivalent to *Microcystis aeruginosa,* Utex# 2835). Fluorescence of phycocyanin was measured using a Turner Design hand-held Fluorometer and a Yellow Springs Instrument Probe (6131 BGA sensor).

- Microcystins: **Cyanotoxins**

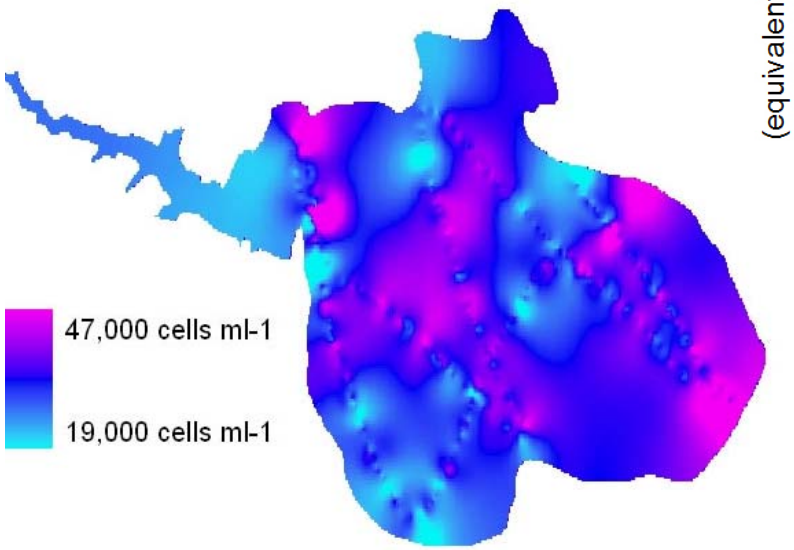
Samples were frozen and thawed in triplicate to lyse cells and concentrated 10-fold by lyophillization. Microcystin concentrations were determined using the Envirologix Quantiplate Kit for Microcystins, Portland, ME (tests results are equivalent to all variants of microcystin and nodularins*). Detection range (with standards) were between 25 and 2500 ng MC L^{-1}, (the lower limit of detection for concentrated water (x10) is therefore 2.5 ng MC

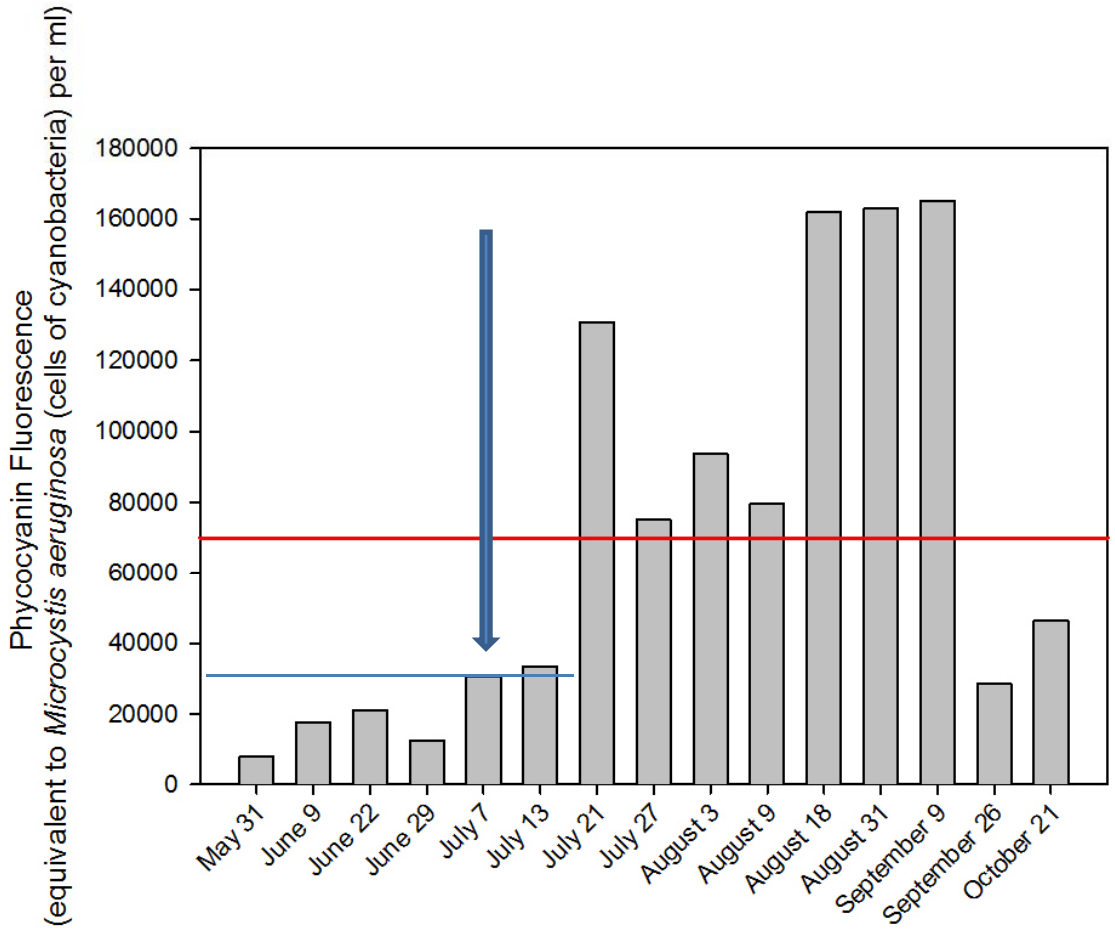
Lake Water Quality Monitoring:

1) Sampling should be done mid-day between the hours of 10 am and 3 pm.
2) In order to obtain a sample that is representative of the entire lake, it is necessary to collect samples from several locations. The number of locations needed depends on the size and complexity of the lake.
3) Samples from each of the locations may be combined for a single toxicity test. (They can also be stored and analyzed separately if information on the spatial variation of microcystin concentrations in the lake is desired..
4) To minimize variability due to vertical strata, collect water by lowering an "integrated tube sampler" to a depth of 3 meters (see attached description of constructing a tube sampler and its operation). Sampling through the metalimnion may be necessary. Take care not to sample close to bottom sediments.
5) Combine samples from all locations into a single large container (e.g. empty 1 gal drinking water bottle).
6) After all samples are collected, shake the collection container thoroughly and pour into the 1 liter sample bottle to approximately ¾ full, leaving space for freezing.
7) Put the sample on ice and in the dark until drop-off at UNH CFB lab.
8) Freeze sample if storage time, prior to delivery, exceeds 12 hours.

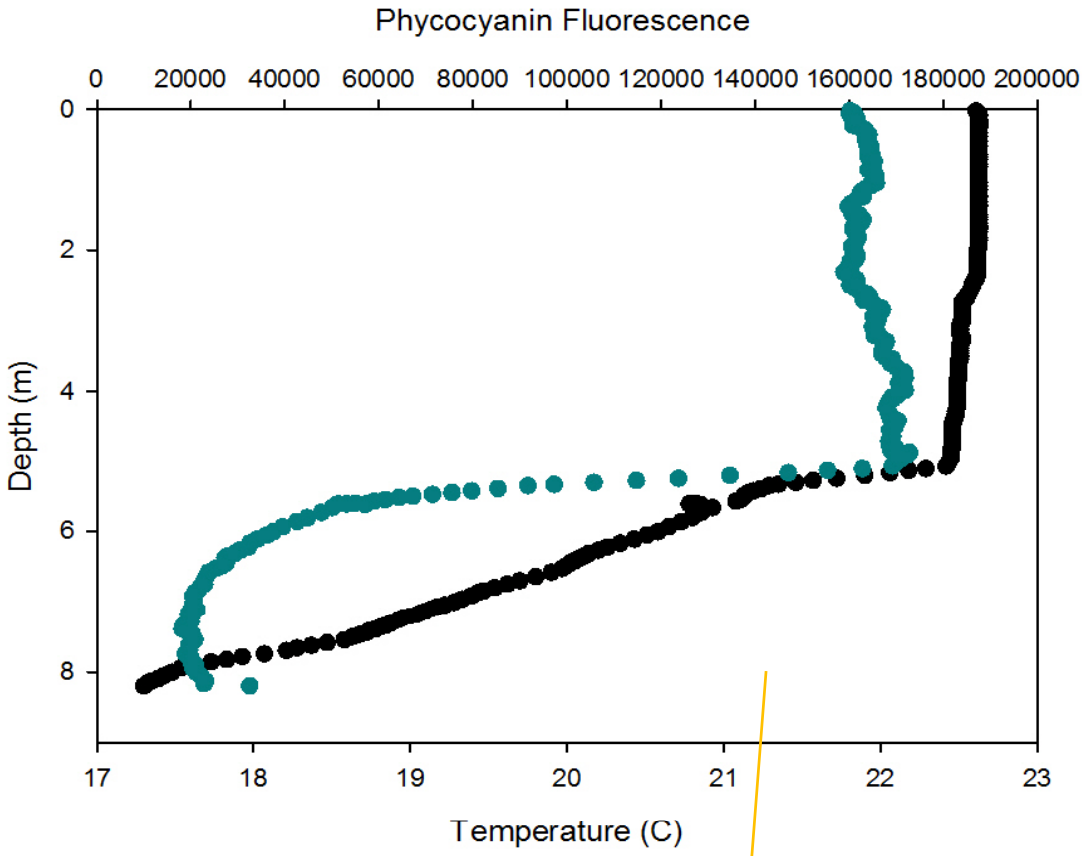
Synoptic lake sampling: Cyanobacteria via PC fluorescence

Lake Attitash July 7, 2010





Spatial variation accounted for
Average phycocyanin values from 5 integrated sites: July 7th example
Sampling was done by the Lake Attitash Association in conjunction with UNH CFB.

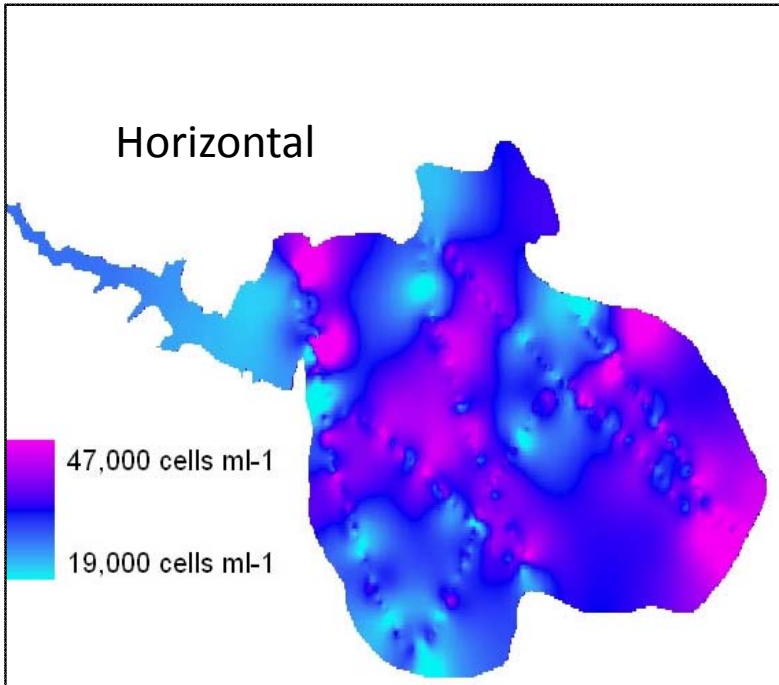


Vertical

Monitoring through Lake Associations (LAA) and education (UNH)

Synoptic lake sampling

Horizontal



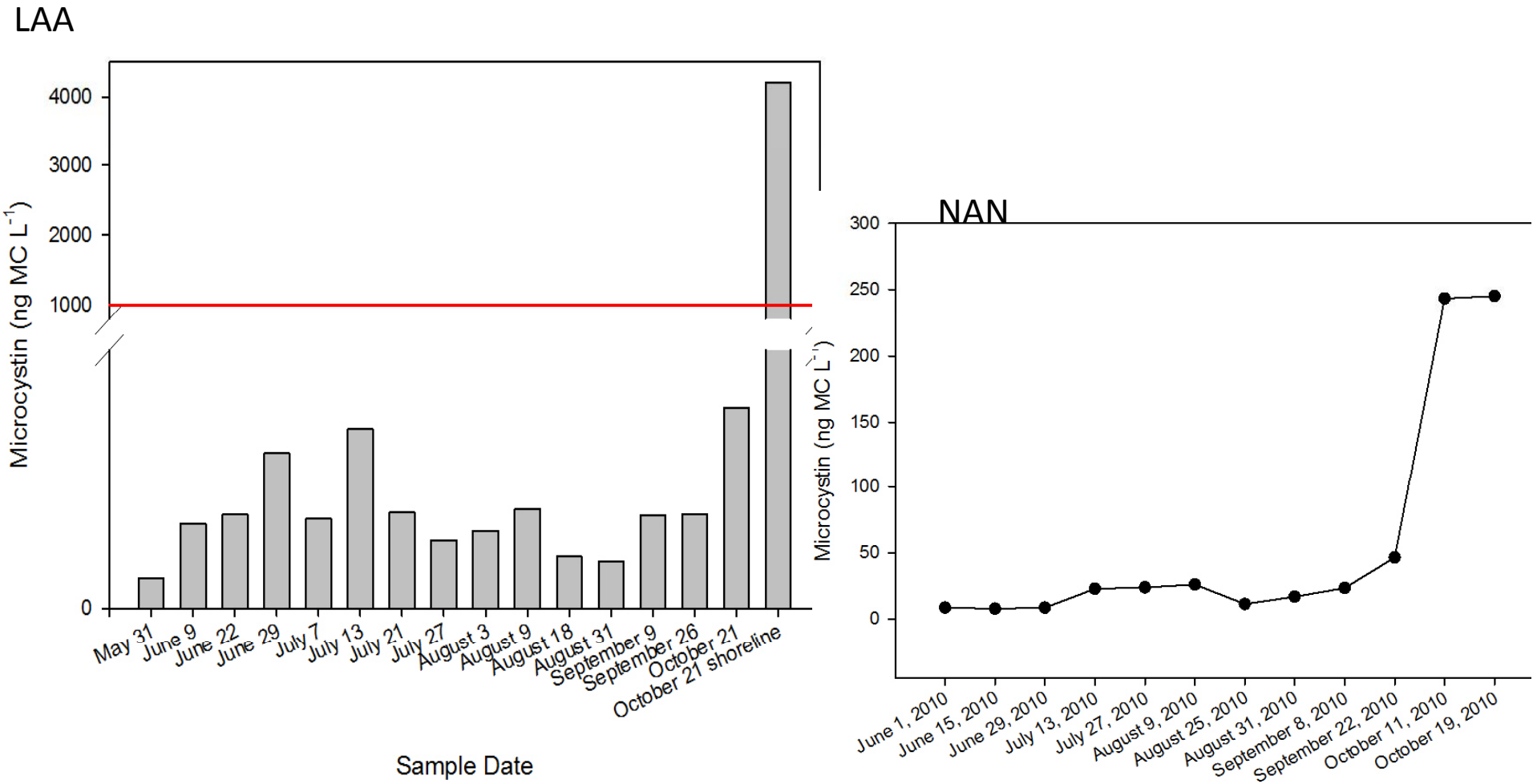
Lake Attitash July 7, 2010

Cyanobacteria Blooms:

1) Visual surface “blooms” of cyanobacteria are important to sample since they often have higher concentrations of cyanobacteria than are present in the open water due to an accumulation effect. If possible, take a picture and submit it electronically in addition to the water sample as it may help identify the dimensions of the bloom.
2) It is recommended that you wear gloves during handling of any cyano-bloom material and wash hands thoroughly when finished sampling.
3) Once the bloom is located, skim the surface with a clean 1 liter HDPE sample bottle to collect the “scum”. Carefully, clean the exterior of the bottle from any scum material.
4) Put the sample on ice until drop-off at UNH CFB lab.
5) Freezing the sample may be necessary if time of drop-off/delivery exceeds 12 hrs.

October Peaks

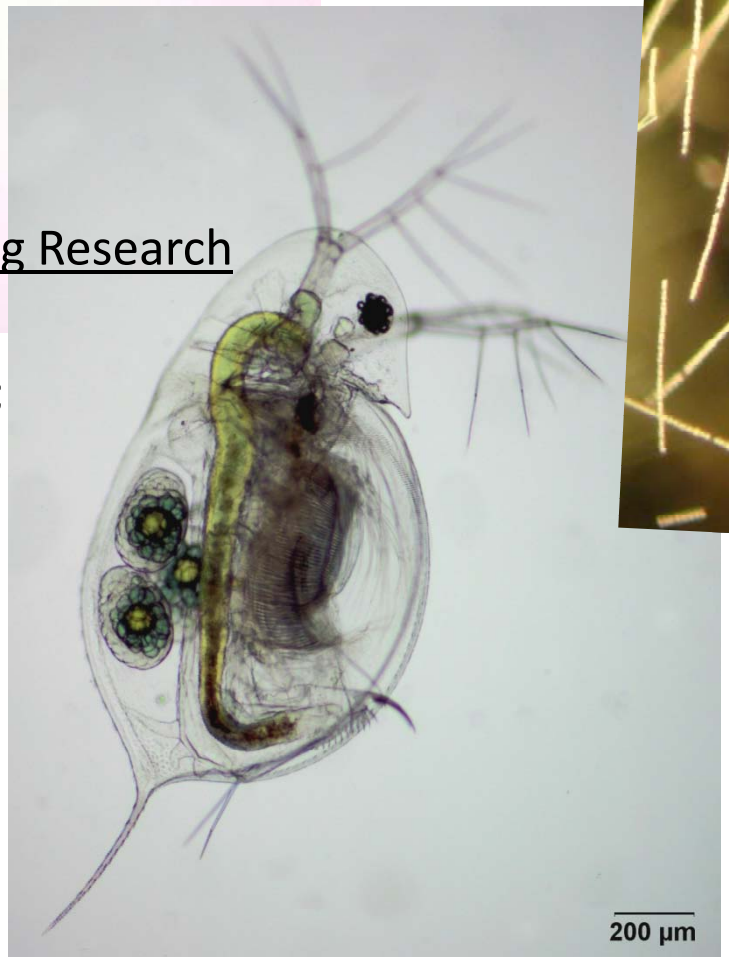
Cyanobacteria abundance and Microcystin toxicity still loom even in the Fall season as far as October or later.



What can long term sampling of cyanobacteria and microcystins tell us about a particular lake?

Could it help explain some of the questions regarding how to monitor, when to sample and which toxins to test for?

How do zooplankton acquire cyanotoxins?



<u>Proposed and on-going Research at UNH:</u>

1. Picocyanobacteria; abundance and toxicity.

2. Cyanotoxins in the food web.

3. Aerosolized cyanotoxins.

Tracking microcystins (MC) in the food web and the potential exposure of MC to humans…