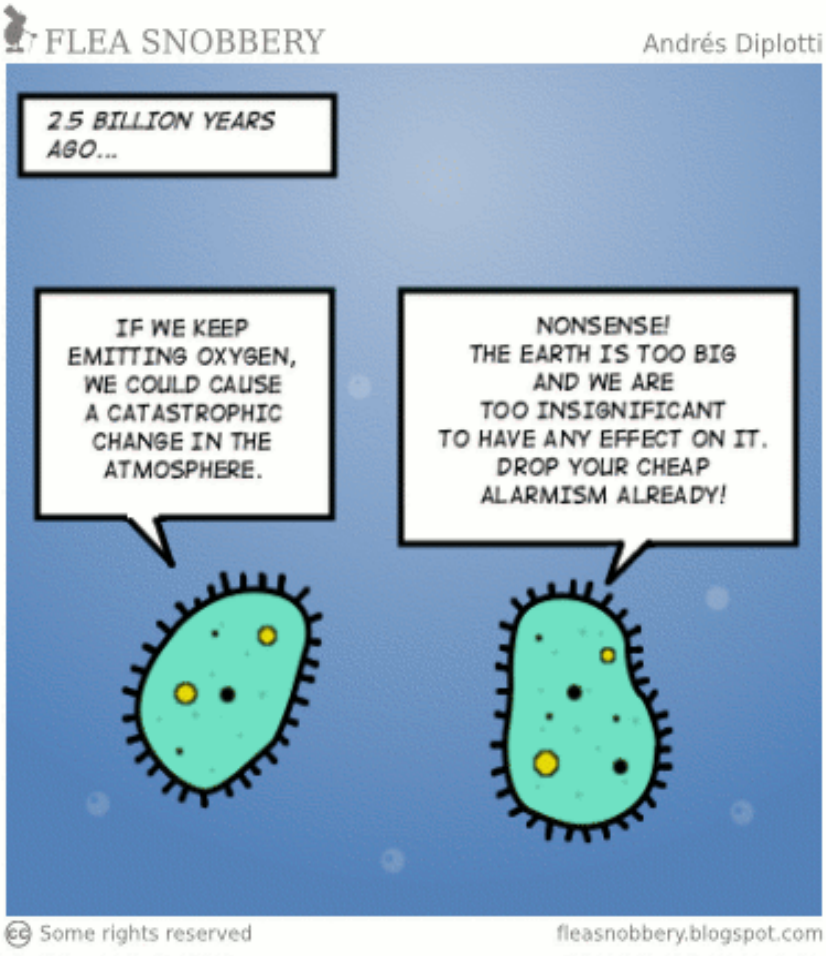
In Vivo Monitoring of Blue-Green "Algae" Using Hydrolab Multi-Parameter Sondes

Patrick A. Sanders
Hach Hydromet
Hydrolab and OTT Products
E-Mail: psanders@hach.com

What are Blue Green "Algae"

- Widely thought to be responsible for creating much of Earth's atmospheric oxygen
- Blue-Green Algae is a common name for the more accurate term: **Cyanobacteria**
- Cyanobacteria are not related to algae, they are actually prokaryotic bacteria
- Utilize chlorophyll based photosynthesis for metabolism
- An important primary producer found in all aquatic systems

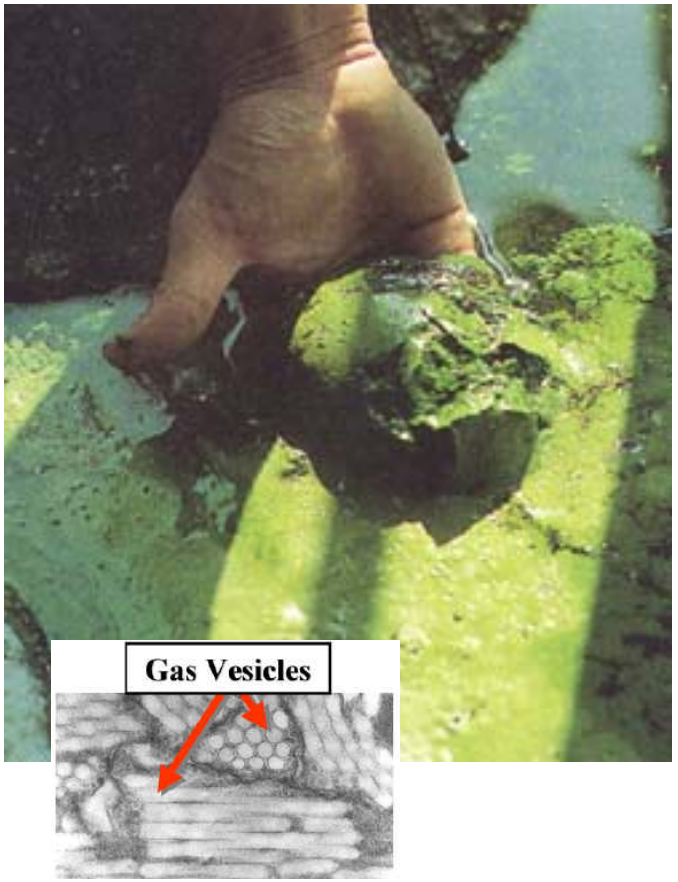


HACH® Hydromet

OTT HYDROLAB®

Cyanobacteria Characteristics

- Can fix molecular nitrogen (N2) from the atmosphere & water and convert it to ammonium
- Nitrogen fixation allows cyanobacteria to maintain high rates of growth
- Can absorb and store excess phosphorus when it is available
- Can adjust their buoyancy to take advantage of better environmental conditions
- Are “hard to handle” and can “taste bad” to primary consumers / grazers



HACH® Hydromet

OTT HYDROLAB®

Why Monitor Cyanobacteria?

- Some species can release neurotoxins and hepatotoxins that can be a public health risk
- Listed by US EPA as a Water Contaminant Candidate
- Blooms are an indicator of eutrophication and can affect water body aesthetics
- Ecological systems can be adversely affected by cyanobacterial blooms
- Can cause taste & odor problems, as well as an increase in filter run times in drinking water plants
- Research on primary productivity, nutrient cycling, ecosystem dynamics, etc.

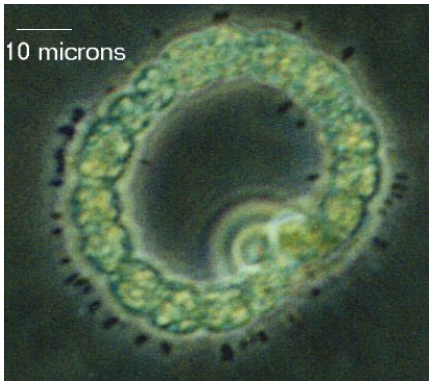
HACH® Hydromet

OTT HYDROLAB®

Noteworthy Cyanobacteria Genera Due to their Ability to Create Cyanotoxins & Form Nuisance Blooms

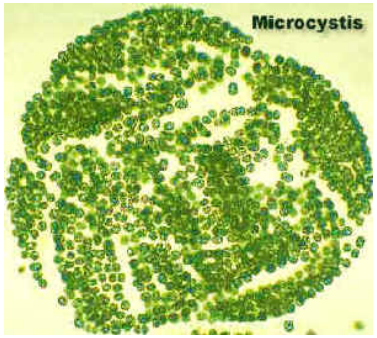
Anabaena spp.

Aphanizomenon spp.



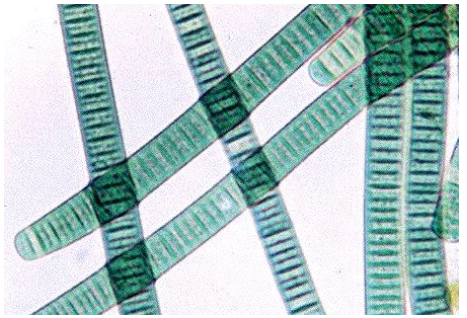
Anabaenaopsis spp.

Cylindrospermopsis spp.



Microcystis spp.

Nostoc spp.



Planktothrix spp.

HACH® Hydromet

OTT HYDROLAB®

Common Cyanobacteria Toxins

Certain genera of Cyanobacteria can produce a wide array of neurotoxins, liver toxins, cell toxins and skin irritants. Currently ~46 species of Cyanobacteria have been shown to be toxic to vertebrates (Chorus & Bartrum 1999)

Toxin	Cyanobacteria Genera
Microcystin (affects liver)	*Microcystis, Anabaena, Planktothrix, Nostoc*, *Anabaenopsis*, & more
Anatoxin-a (affects nerve synapse)	*Anabaena, Aphanizomenon*
Cylindrospermopsin (affects liver)	*Cylindrospermopsis, Aphanizomenon*
Saxitoxins (affects nerve axons)	*Anabaena, Cylindrospermopsis, Aphanizomenon, &* more
Lipopolysaccharides (potential irritant, affects exposed tissue)	All

HACH® Hydromet

OTT HYDROLAB®

Top 6 Ways to Assess Whether a Cyanobacterial Problem Exists or is Likely

From the World Health Organization (1999)

Table 2: Information to assess whether a cyanobacterial problem exists or is likely, from WHO 1999.

Observation	Sources of Information	Management Options
1. Potential for bloom formation	Water quality monitoring data (nutrients, temperature, etc)	Basis for proactive management
2. History of bloom formation	Cyanobacterial blooms may follow marked seasonal and annual patterns	Can inform proactive management
3. Monitoring for Cyanobacteria and/or cyanotoxins	Turbidity, discoloration, cell microscopic identification, cell counts and toxin analysis provide increasingly reliable information	Possible only during event and enables only reactive management
4. "Scum Scouting"	In areas of high public interest the general public and untrained agency staff may play a role in identifying and reporting obvious hazards such as scums	Possible only during event and enables only reactive management
5. Reporting of animal deaths and human illness	Requires both volition and a mechanism for data collection which may not exist	Possible only during event and informs only reactive management
6. Epidemiological detection of disease patterns in the human population	Requires both effective reporting and large scale effects before detection is likely	Normally well after an event; can inform future management strategies

Source: World Health Organization, 1999

In Vivo Fluorometry (IVF) can Enable Proactive Management of Cyanobacteria

In Vivo

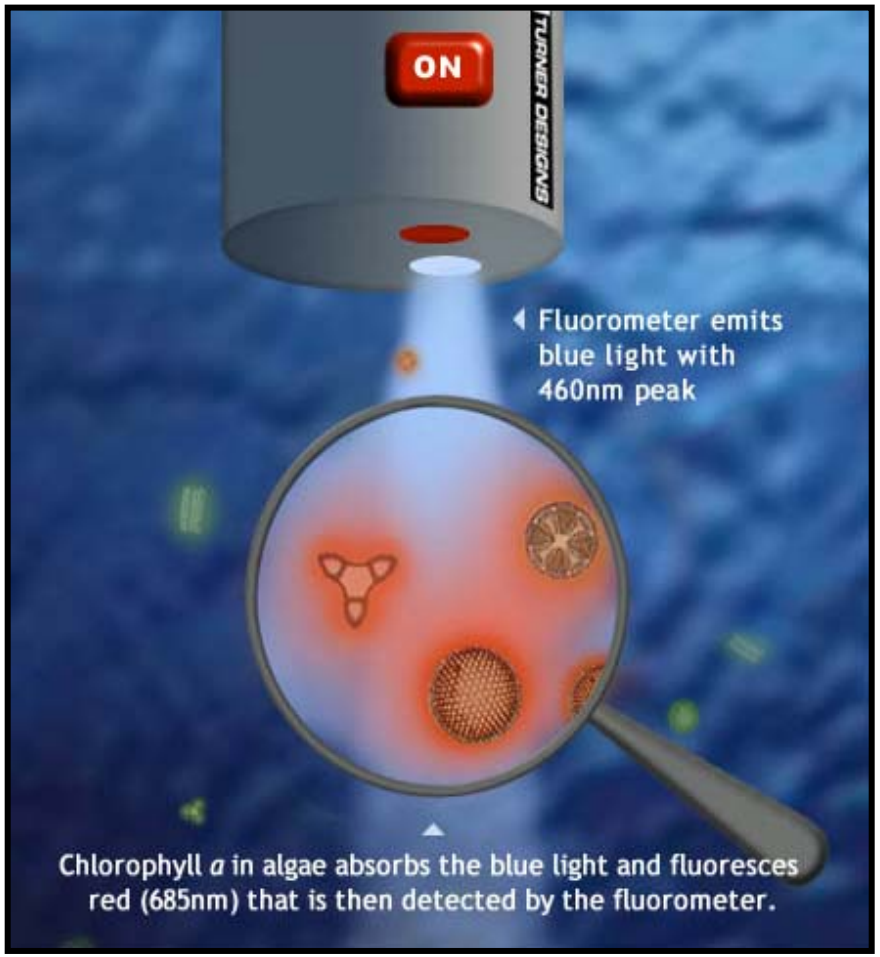
- Occurring or made to occur within a living organism or natural setting

Fluorescence

- The phenomena of some compounds to absorb specific wavelengths of light and instantaneously emit longer wavelengths of light energy

In Vivo Fluorometry Benefits

- Ability to collect large quantities of data quickly & easily, w/o special handling
- Ability to collect & view data in real-time
- Ability to build up historical datasets



graphic from Turner Designs – used w/ permission

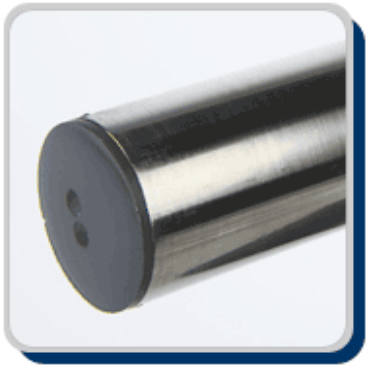
HACH® Hydromet

OTT

HYDROLAB®

Using In Vivo Fluorescence on a Hydrolab for Cyanobacteria Monitoring

- Hydrolab sondes can be used for multiple water quality parameters, including cyanobacteria.
- Cyanobacteria is measured using *in vivo* fluorescence sensors manufactured by Turner Designs.
- IVF sensor characteristics include:
  - high sensitivity and low noise
  - fast response
  - wide dynamic range
  - excellent turbidity rejection
  - high quality fluorescence data



HACH® Hydromet

OTT HYDROLAB®

What is being Measured with our Sensors?
Chlorophyll a / Phycocyanin / Phycoerythrin

<u>Chlorophyll *a*</u> (chl *a*) (Ex: 440nm / Em: 680nm)

- Located within both photosystems (PS I and PS II) in phytoplankton chloroplasts and in cyanobacteria
- Absorbs light for photosynthesis
- Used to estimate phytoplankton biomass

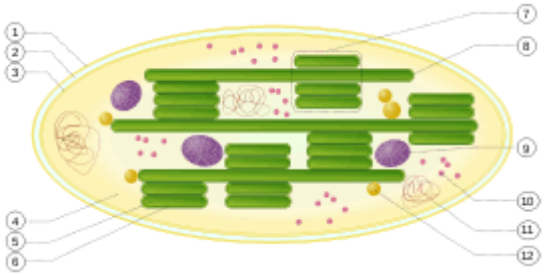


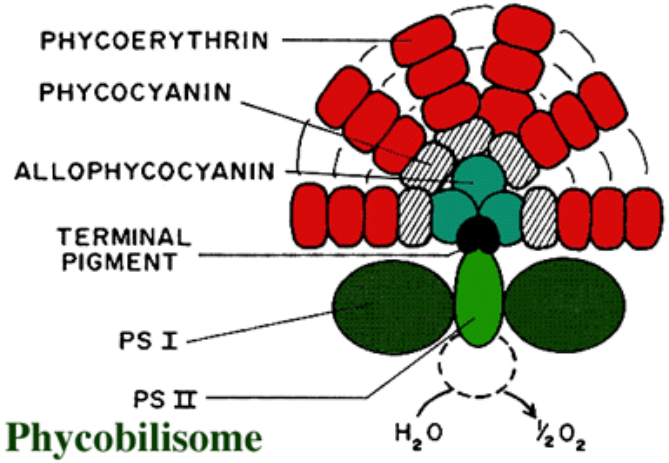
Image source: Wikipedia

<u>Phycocyanin</u> (PC) (Ex: 595nm / Em: 650nm)

- Used primarily for freshwater BGA applications
- Light harvesting antennae of PS II

<u>Phycoerythrin</u> (PE) (Ex: 525nm / Em: 570nm)

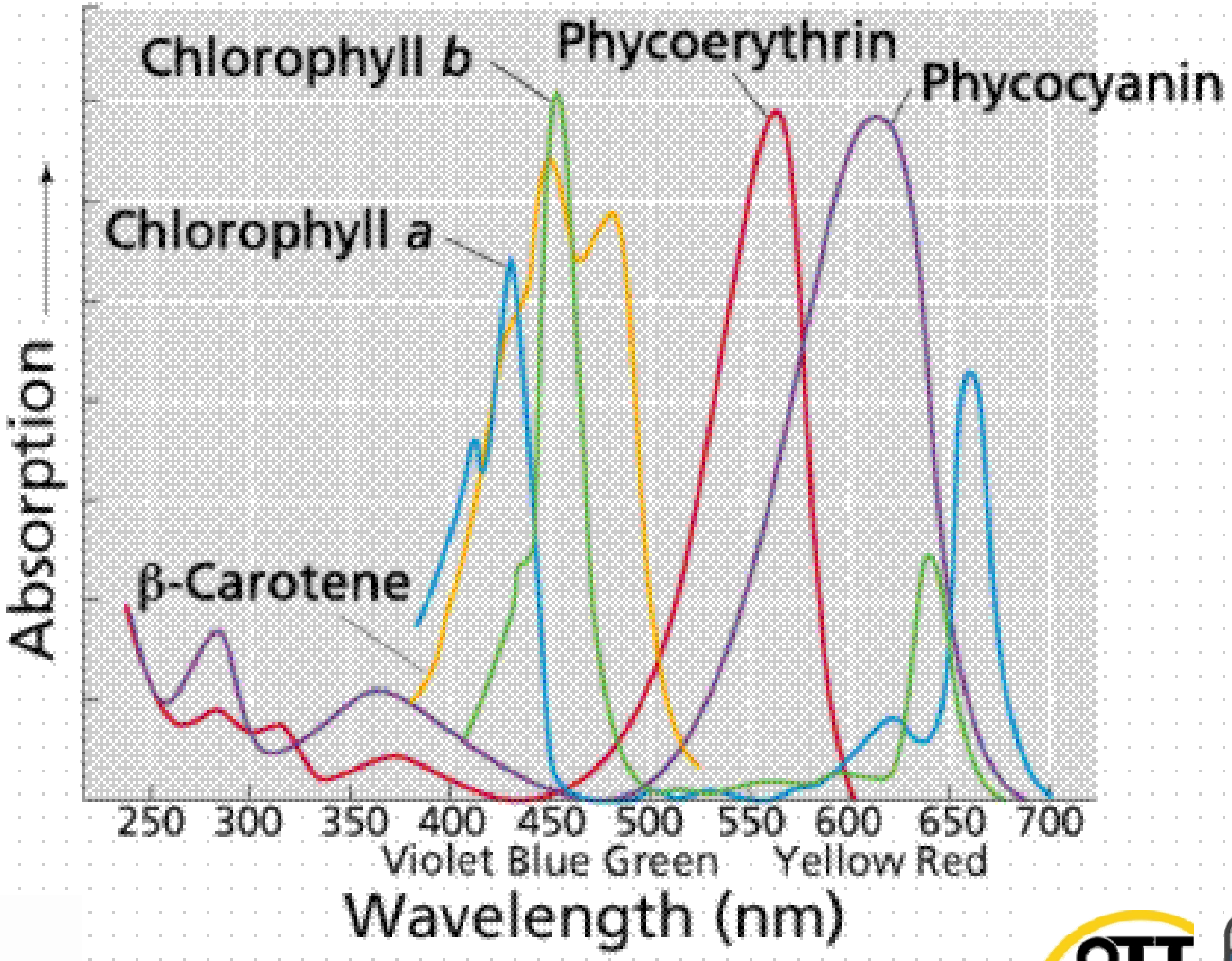
- Used primarily for marine BGA applications
- Light harvesting antennae of PS II



HACH® Hydromet

OTT HYDROLAB®

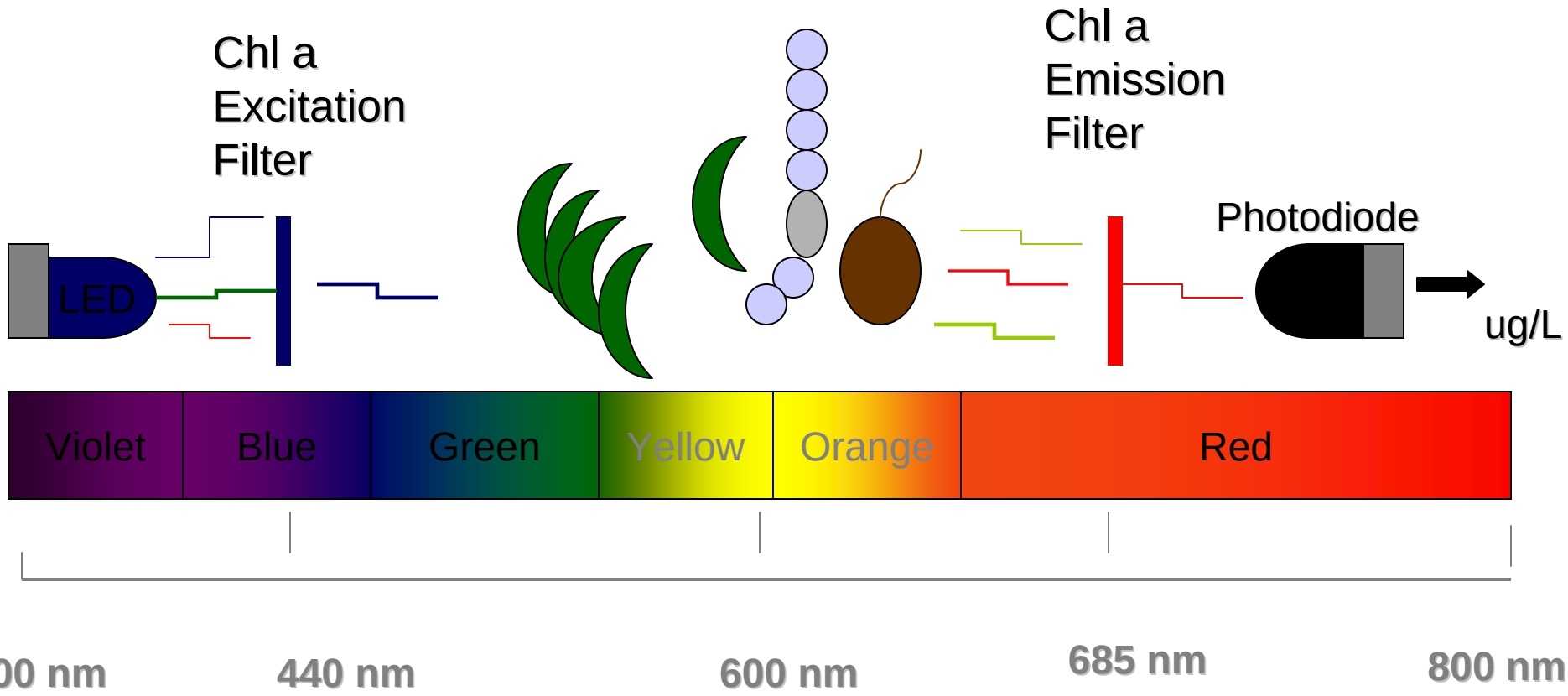
Absorption Spectra for Chl a, PC, & PE



HACH® Hydromet

OTT HYDROLAB®

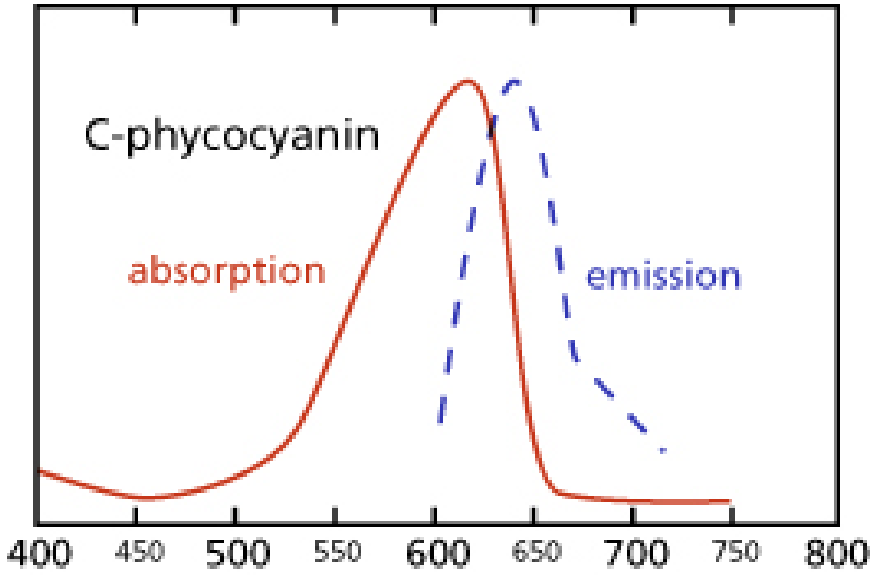
Optical Configuration for Chlorophyll a

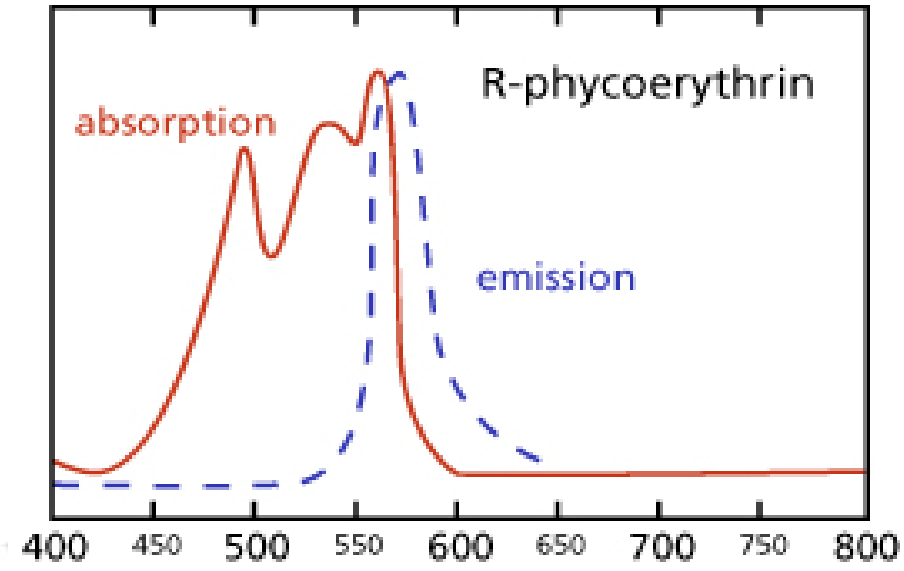


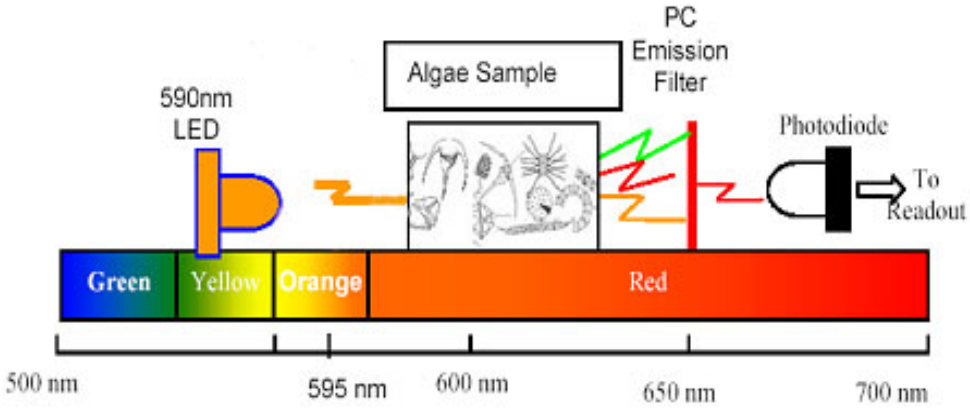
HACH® Hydromet

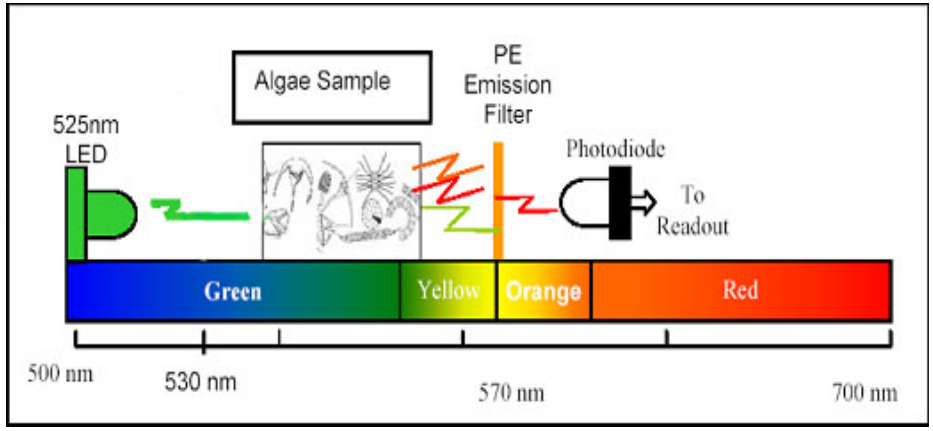
OTT HYDROLAB®

Optical Configurations for Phycocyanin & Phycoerythrin









HACH® Hydromet

OTT HYDROLAB®

Calibration Standards & Data Correlation

- Adjustable solid secondary standard accessory available for calibration and to check for sensor stability and drift.

- Sensor data correlations can be made with:
  - cell counts
  - extracted pigments (HPLC, absorbance)
  - presence of taste and odor
  - filter run times

- Purified Phycobilin Pigments Available:
  - Prozyme (www.prozyme.com)

HACH® Hydromet

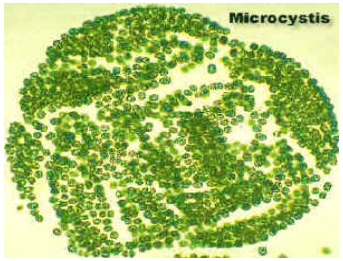
OTT HYDROLAB®

Sensor Performance

Phycocyanin

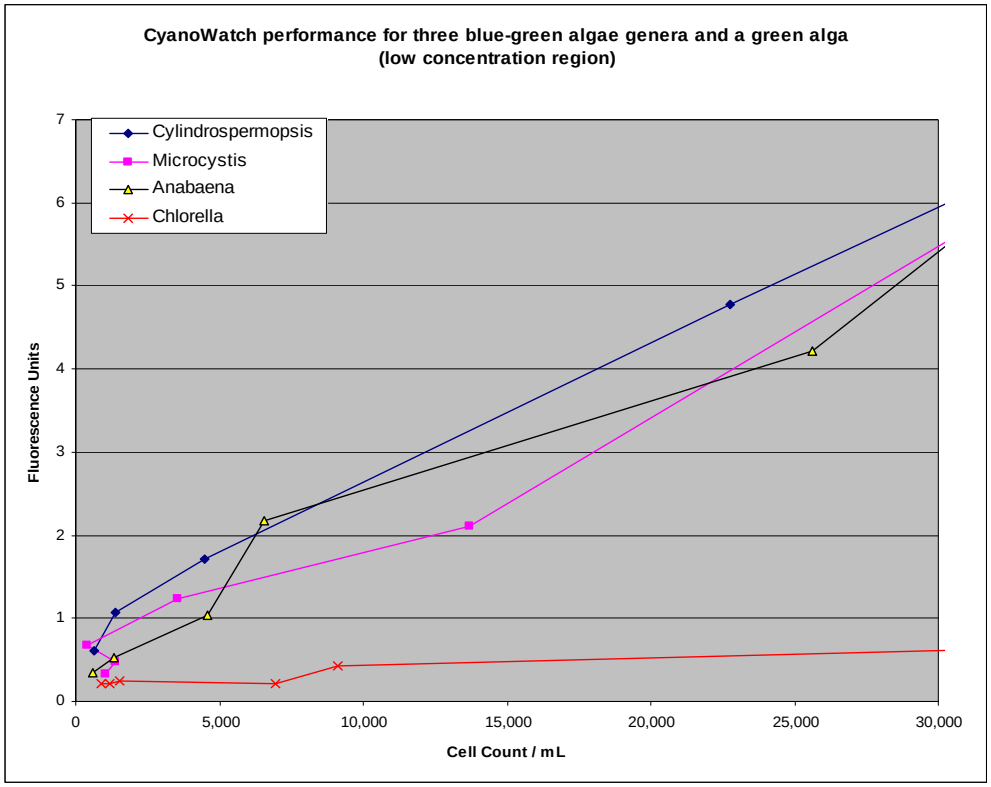
•Ex 595nm / Em 650nm

Cylindrospermopsis spp.



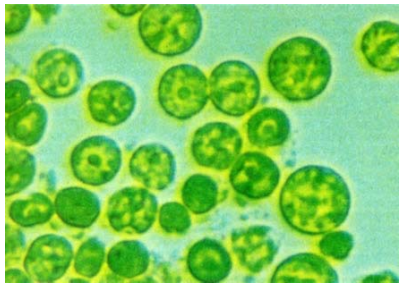
Microcystis spp.

Anabaena spp.

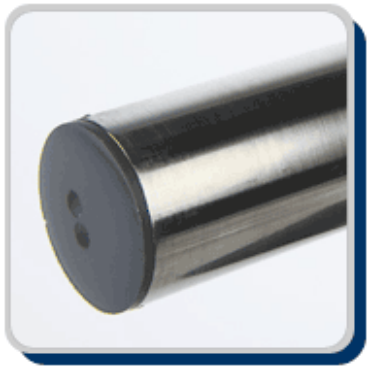


Chlorophyll a

•Ex 440nm / Em 680nm

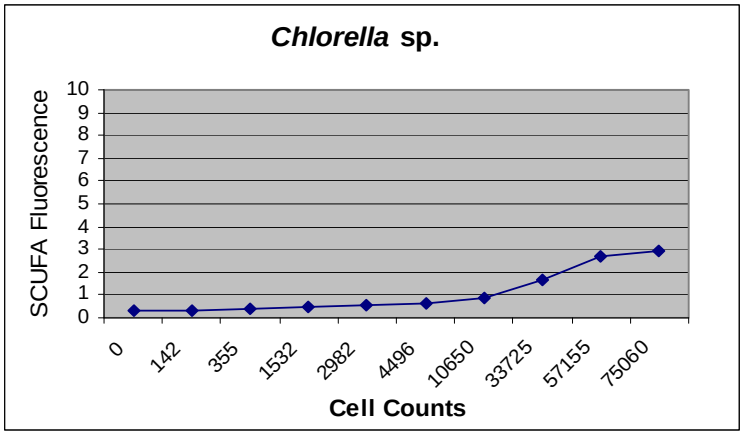


Chlorella spp.



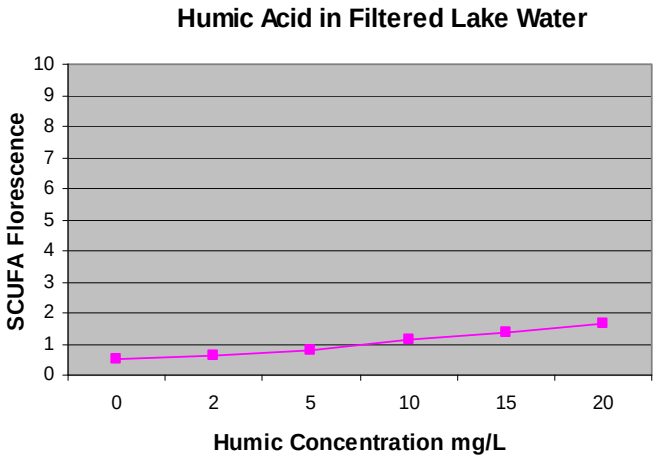
Interfering Substances

Green Algae



- Algal groups not containing phycocyanin do not interfere with fluorescence readings

Humic Acid

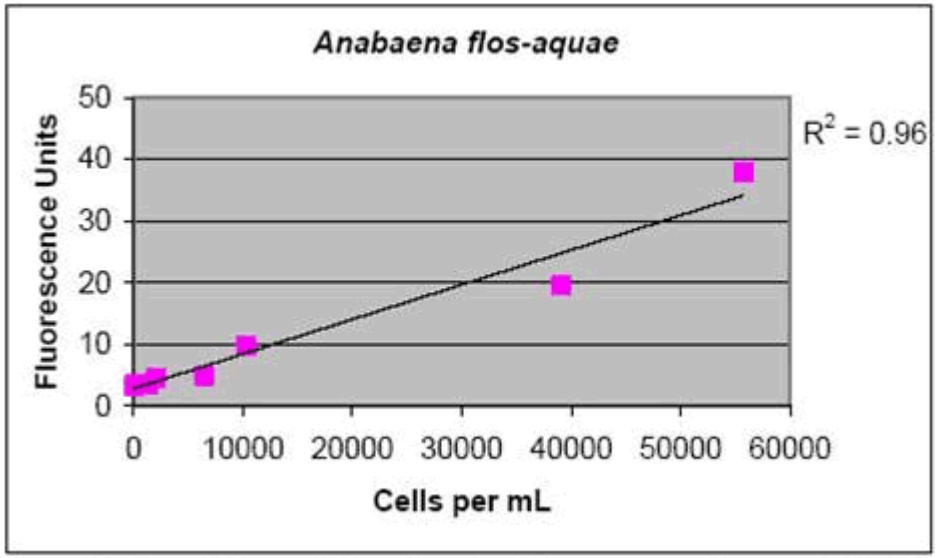


- At high humic concentrations there is no significant fluorescence signal indicating that the optics are susceptible to humic interference

HACH® Hydromet

OTT HYDROLAB®

Example: Anabaena flos-aquae IVF Correlation to Cell Counts



Minimum Detection Limit: ~150 cells/mL

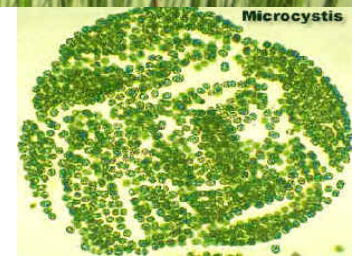
Linear up to >150,000 cells/mL

Accuracy +/- 3% for signal levels > 0.5% FS

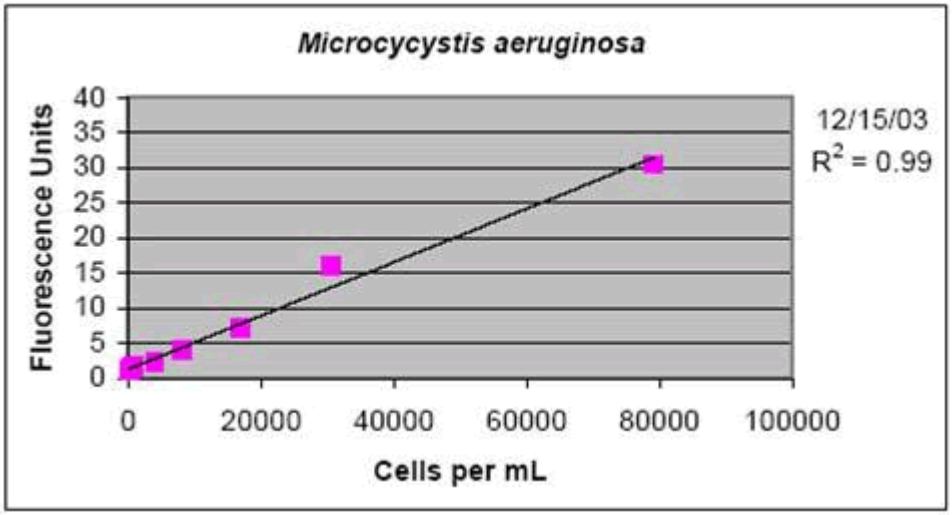
HACH® Hydromet

OTT HYDROLAB®

Example: Microcystis aeruginosa IVF Correlation to Cell Counts



Microcystis spp.



Minimum Detection Limit: ~150 cells/mL

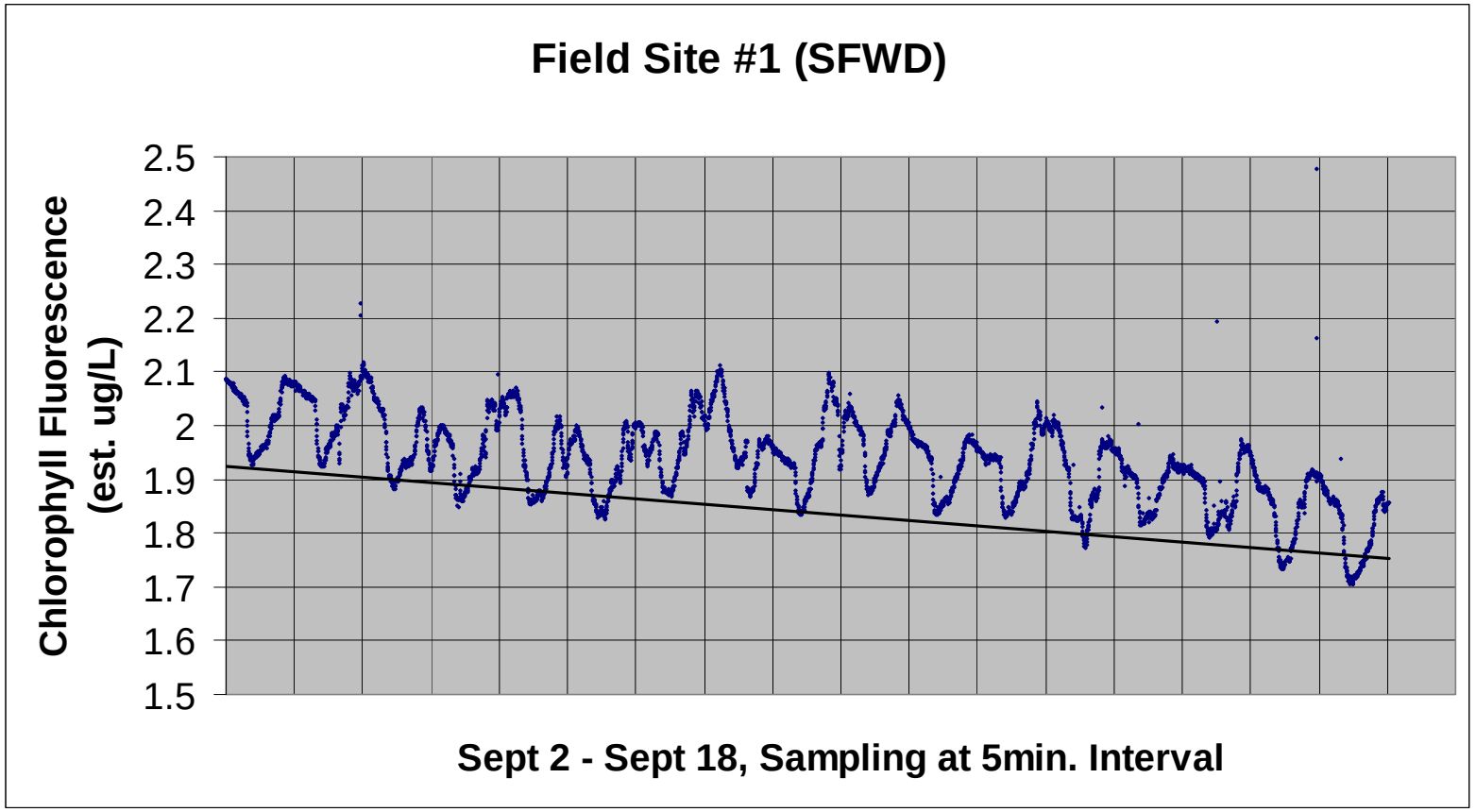
Linear up to >150,000 cells/mL

Accuracy +/- 3% for signal levels > 0.5% FS

HACH® Hydromet

OTT HYDROLAB®

The Value of Real-Time IVF Data



HACH® Hydromet

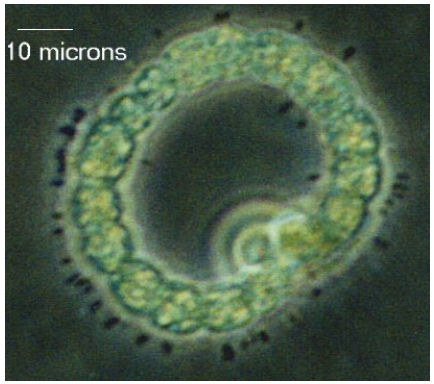
OTT HYDROLAB®

Anabaena spp.

Anabaena spp.

Capable of producing the hepatotoxin Microcystin and the neurotoxins Anatoxin-a and Saxotoxin.

Anabaenopsis spp.



Anabaenaopsis spp.

Capable of producing the hepatotoxin Microcystin.

Aphanizomenon spp.

Aphanizomenon spp.

Capable of producing the hepatotoxin Cylindrospermopsin, and the neurotoxins Anatoxin-a and Saxotoxin.

HACH® Hydromet

OTT HYDROLAB®

Cylindrospermopsis spp.

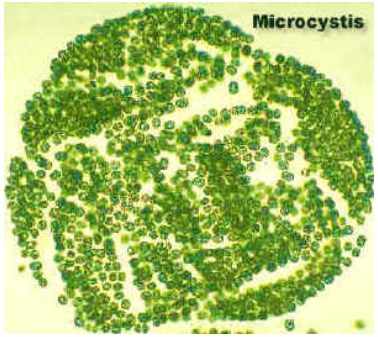
Cylindrospermopsis spp.

Capable of producing the hepatotoxin Cylindrospermopsin, and the neurotoxin Saxotoxin.

HACH® Hydromet

OTT HYDROLAB®

Microcystin spp.



Microcystis spp.

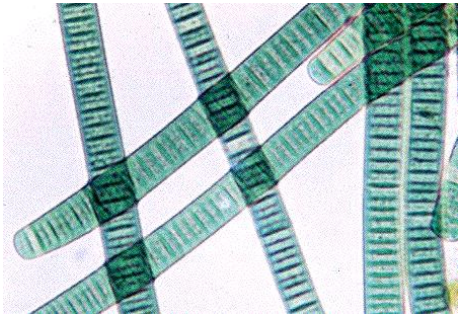
Capable of producing the hepatotoxin Microcystin.

Nostoc spp.

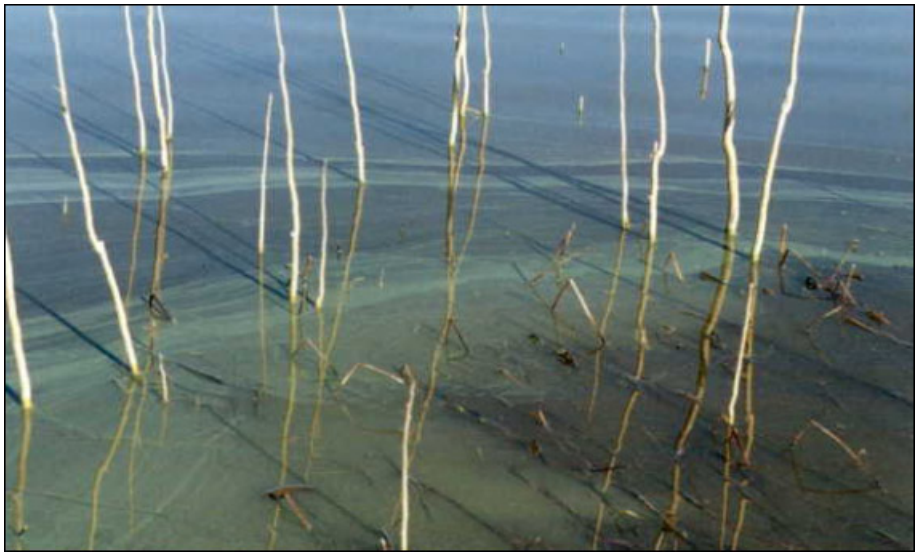
Nostoc spp.

Capable of producing the hepatotoxin Microcystin.

Planktothrix spp. (Oscillatoria)



Planktothrix spp.



Capable of producing the hepatotoxin Microcystin.

Thank you for your attention.
Questions?