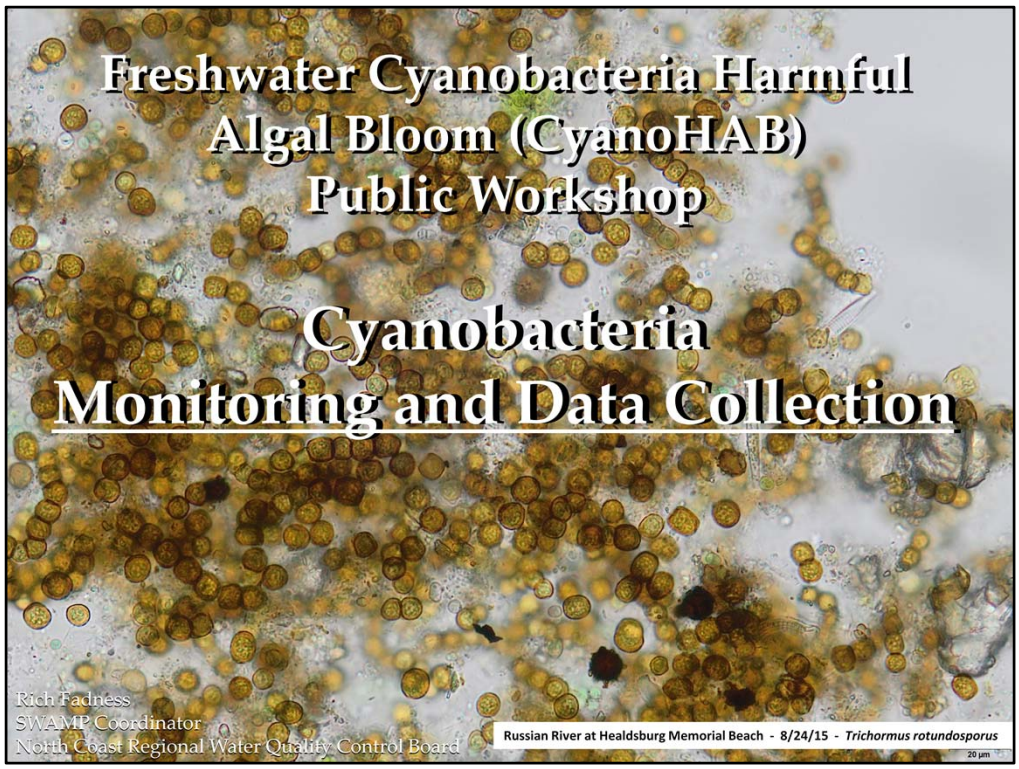
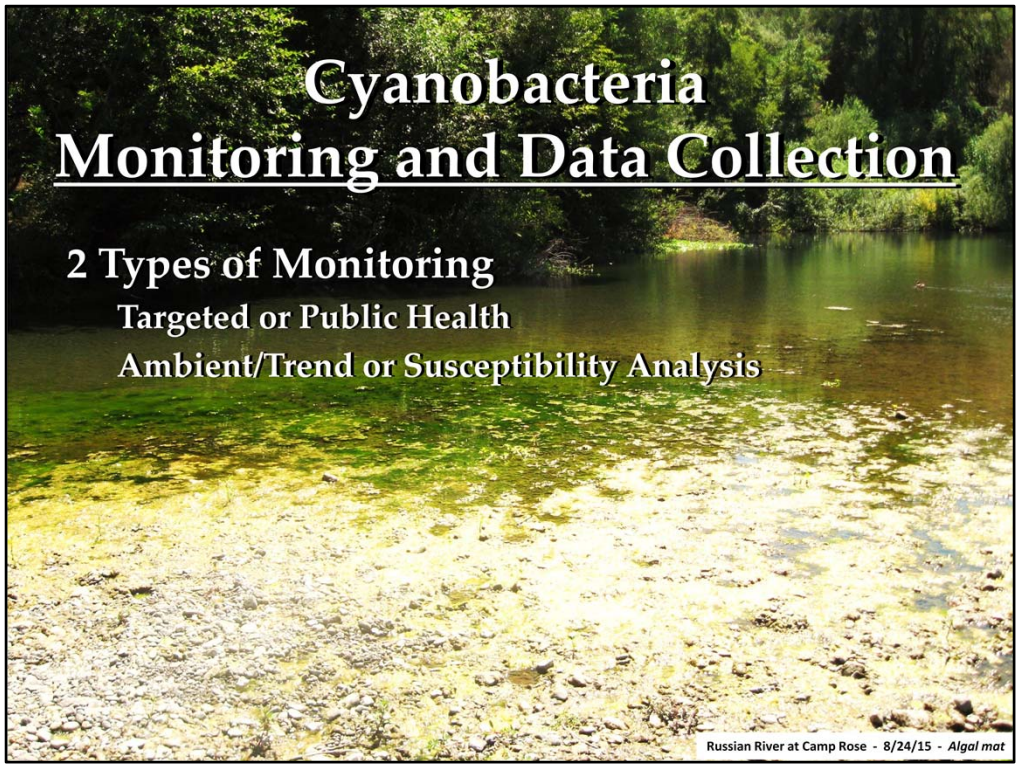
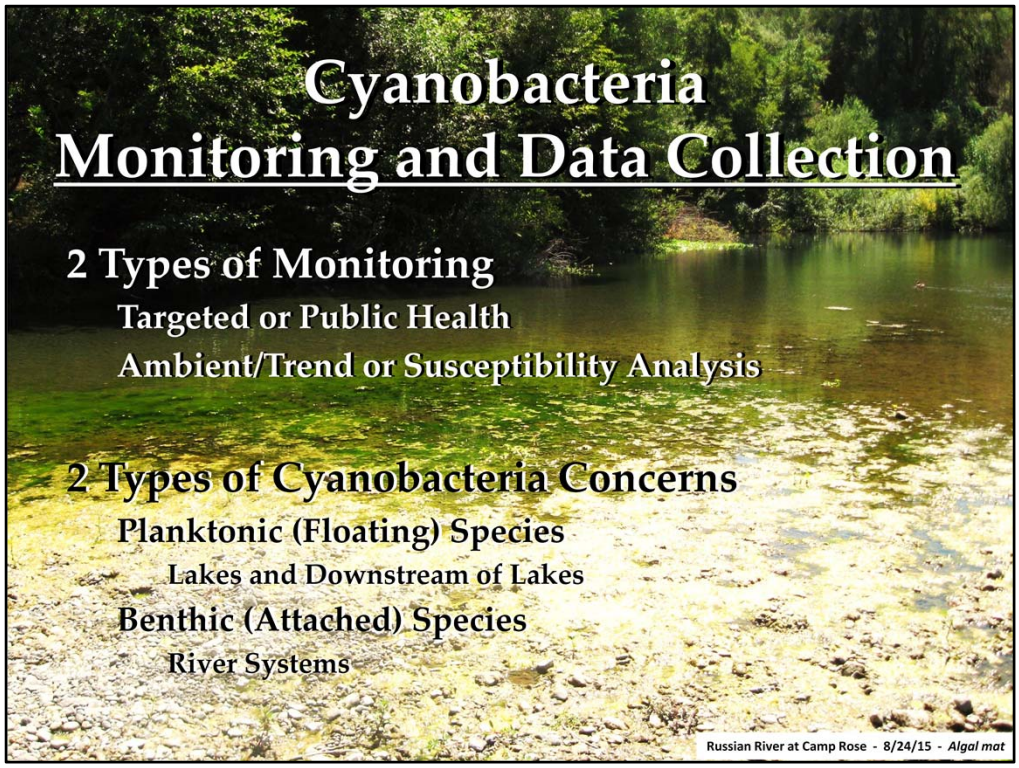

Freshwater Cyanobacteria Harmful Algal Bloom (CyanoHAB) Public Workshop
Cyanobacteria Monitoring and Data Collection
Rich Fadness
SWAMP Coordinator
North Coast Regional Water Quality Control Board
Russian River at Healdsburg Memorial Beach - 8/24/15 - Trichormus rotundosporus
20 µm



Today I'm going to focus on 2 types of monitoring:

1) Targeted or Public Health monitoring – Here we focus on the protection of people, their pets and livestock. This is what really brings us together today.

and 2) Ambient or Trend Monitoring for what I call "Susceptibility Analysis". To determine the susceptibility of a waterbody to develop a bloom, we need to collect the relative information and ambient water quality data that will assist us in further understanding the dynamics or characteristics of each waterbody. What are the conditions or drivers of cyanobacteria bloom development that we can use to assist us when determining if and when sampling is necessary.



As part of these monitoring efforts we need to be aware of and concerned with the 2 types of cyanobacteria populations, planktonic and benthic.

Planktonic are the free-floating species most commonly associated with lakes or rivers which drain lakes. These are the cyanobacteria that color the water with various shades of green and end up capturing the headlines on the news. They affect large waterbodies, turning lake water green and covering beaches and coves with scums of multi-colored hues. These are the cyanobacteria for which most monitoring and response plans have been written to date.

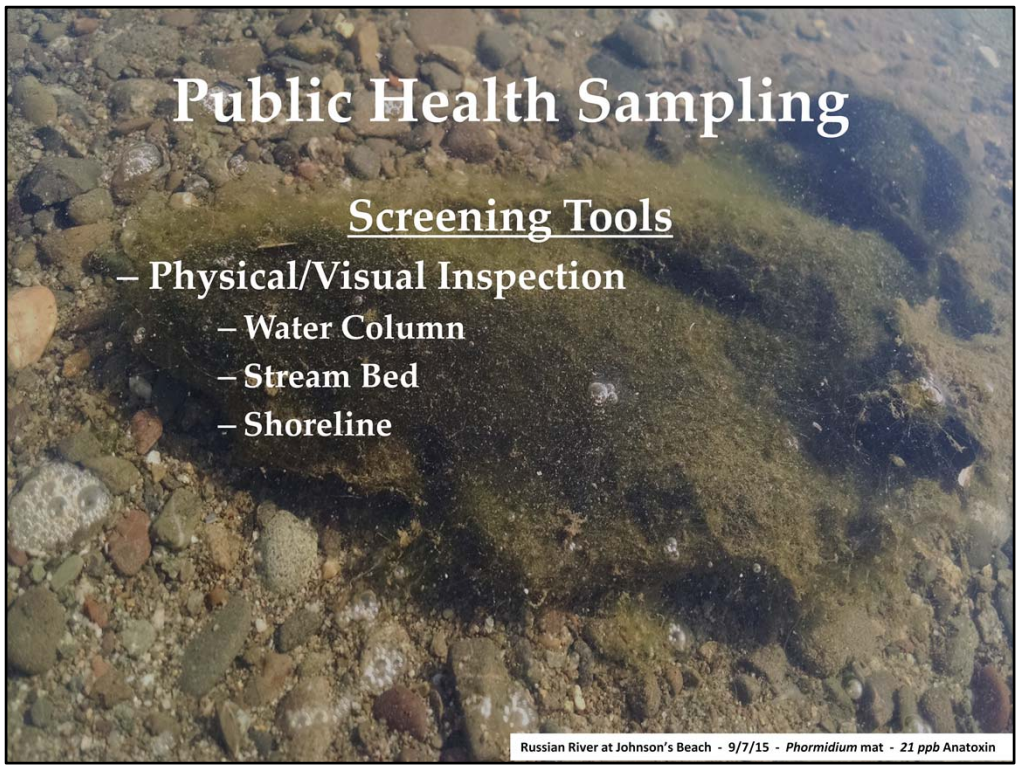
Benthic or attached species on the other hand are predominantly found in rivers and streams and tend to be rather non-descript and even hidden from sight. A river can have clear running water and yet be toxic due to the cyanobacteria that clings to the streambed releasing its toxins.

Later this afternoon, we will hear of some case studies that demonstrate both of these situations.

With regards to cyanobacteria sampling, targeted sampling is driven by the need to protect Public Health and is therefore focused on providing the information necessary to assist Resource Managers and Public Health Departments in making the decisions on whether or not a lake or river should be posted or even closed to recreational use.

These decisions have affect on all forms of recreation, including swimming, fishing due to bioaccumulation of cyanotoxins, boating including kayaking, skiing and jet-skiing, as well as taking our dogs on a play day or watering livestock.

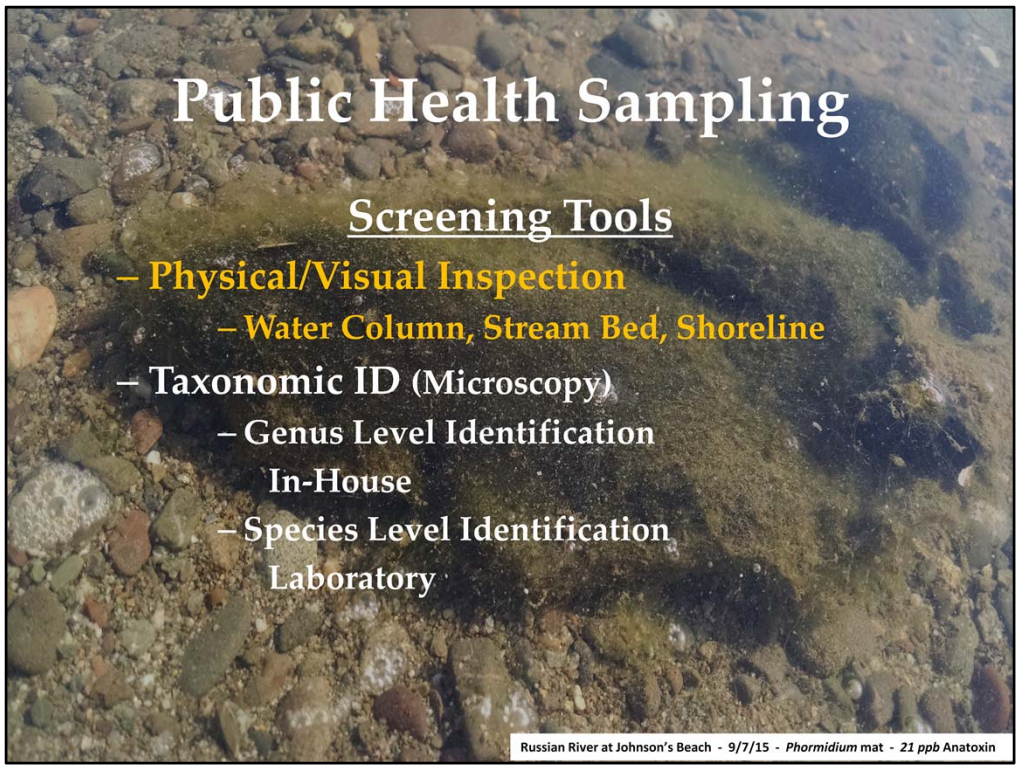
In addition, the information that we collect is extremely important for potable water users, both municipal and private. In many locations throughout the Region, urban and independent users draw their water supply from the rivers of the north coast.



Sampling for cyanotoxins on a regular or ongoing basis is an expensive endeavor. We need to find ways to determine when sampling is appropriate and necessary. For that we can use a variety of screening tools and environmental triggers to assist us with that determination (susceptibility analysis).

Initially, we can make visual observations of the water column, streambed, or along the shorelines of our rivers and lakes.

Is there suspended or planktonic cyanobacteria in the water column, attached or benthic cyanobacteria on the streambed or accumulations of cyanobacteria mats or scums present in the nearshore beach area or along the shoreline.



Another screening tool of some effectiveness is taxonomic identification. Not all genera of cyanobacteria produce cyanotoxins, but many do. Knowing how to identify the dominant genera within a waterbody can assist in determining when sampling should begin.

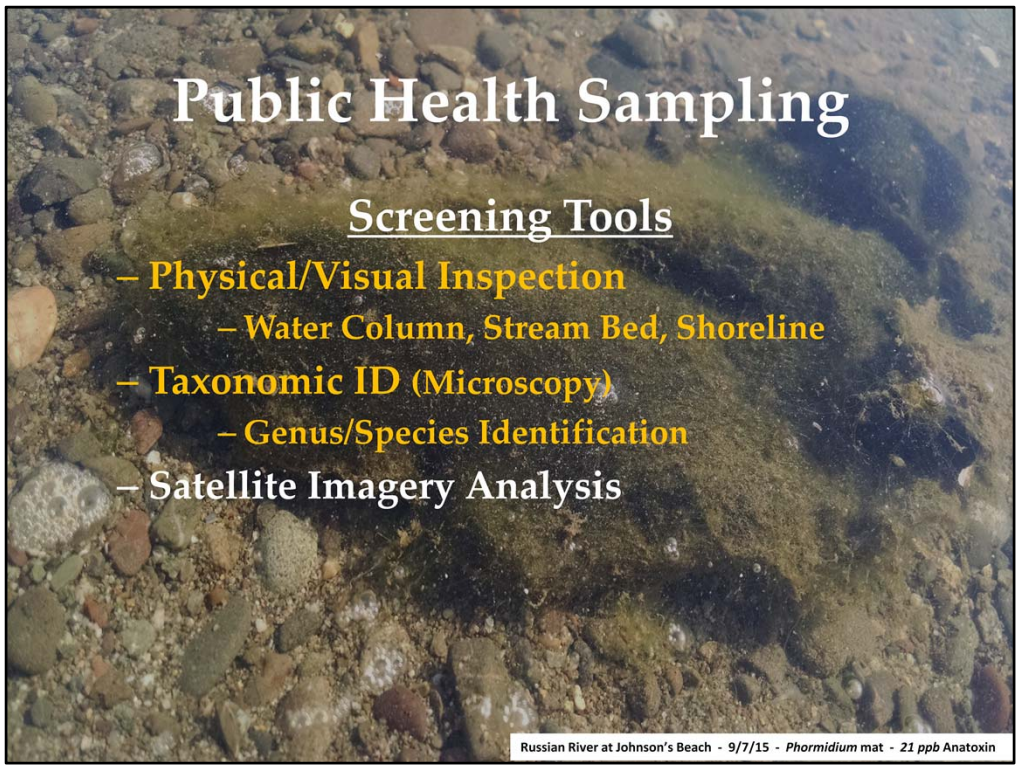
A confounding factor in using taxonomic ID as a tool is that not every species within a genera is capable of producing toxins, and even known toxic species may or may not produce toxins at any given time.

With training, cyanobacteria identification to the genus level may be accomplished in the field or under a microscope but individual species identification is best handled by professional taxonomists.

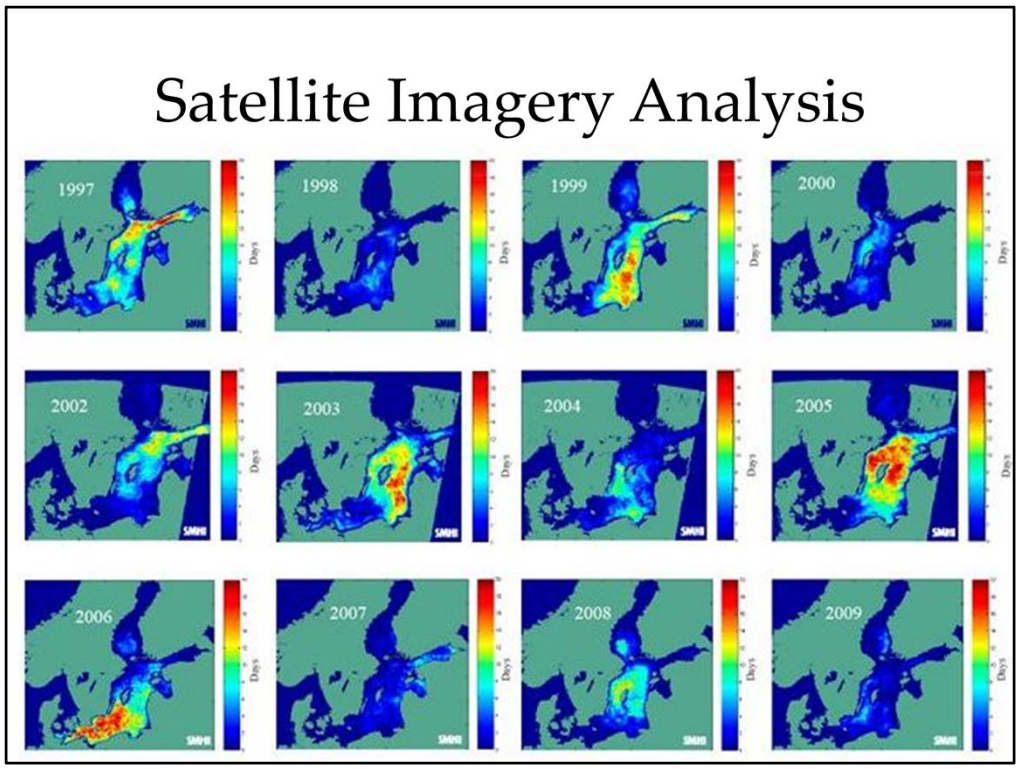
Taxonomic Identification

Observed Genera w/Toxic Species	Possible Toxin(s)	Observed Genera w/Non-Toxic Species
Anabaena	MICROCYSTINS, ANATOXIN, CYLINDROSPERMOPSIN, SAXITOXIN	*Anabaena*
		Aphanocapsa
Aphanothece	MICROCYSTINS, CYLINDROSPERMOPSIN	*Aphanothece*
Calothrix	MICROCYSTINS	*Calothrix*
		Chroococcus
Cylindrospermum	MICROCYSTINS, ANATOXIN, CYLINDROSPERMOPSIN	
Dolichospermum	MICROCYSTINS, ANATOXIN	
Geitlerinema	MICROCYSTINS, SAXITOXINS	*Geitlerinema*
Gloeotrichia	MICROCYSTINS	*Gloeotrichia*
		Leptolyngbya
Lyngbya	LYNGBYATOXIN	*Lyngbya*
		Microchaete
Nodularia spumigena	NODULARIAN, MICROCYSTINS	
Nostoc	MICROCYSTINS	
Oscillatoria	MICROCYSTINS, ANATOXIN, CYLINDROSPERMOPSIN	
Phormidium	ANATOXIN	*Phormidium*
Pseudanabaena	MICROCYSTINS	
		Spirulina
Trichormus	MICROCYSTINS	*Trichormus*

This table is a list of cyanobacteria species collected from various locations in the Russian River during the summer of 2015. In many cases there were more than one species of each genus...some which are suspected toxin producers while others that are known not to produce toxins. You will notice that some genera are located on both sides of the table.

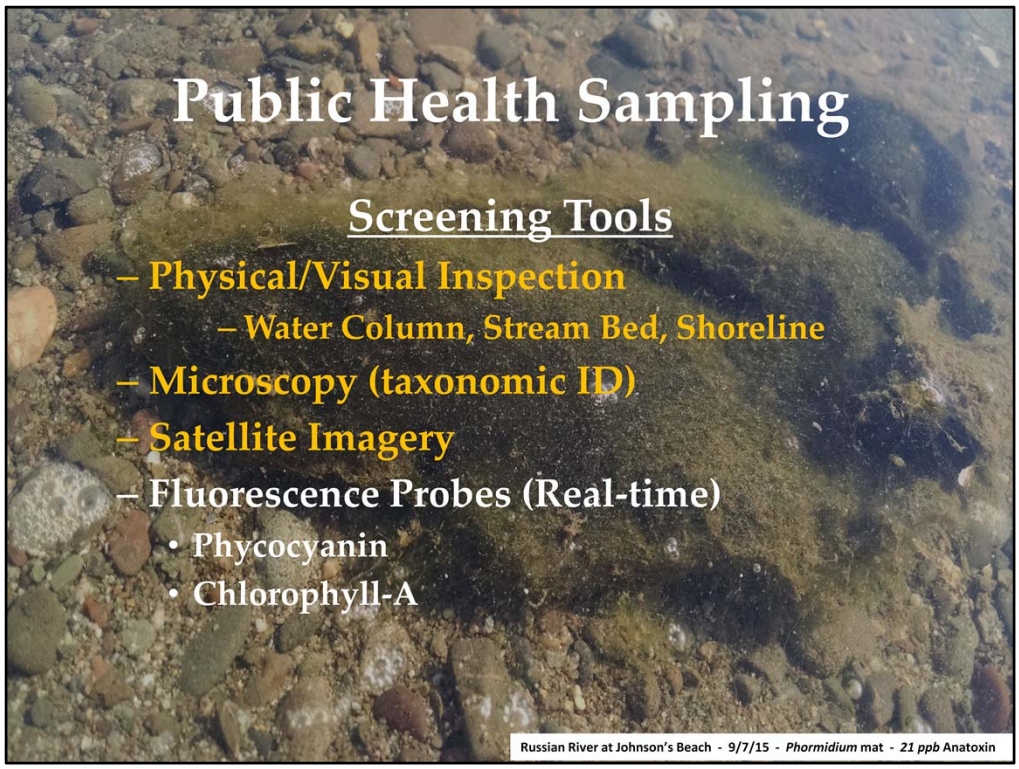


Another screening tool becoming available for use on some of the larger lakes in the region is satellite imagery.



Satellite imagery and analysis provides data on bloom development and extent within the large lakes in the region. This analysis provides information on Cyanobacteria bloom development but does not provide any information on toxin production.

This image is a summary of the number of days in which cyanobacteria was observed in the Baltic Sea in each pixel during the period 1997-2009, based on NOAA-AVHRR satellite imagery. This same type of analysis will provide lake managers with timely data when a bloom is imminent.



In addition, another cost-effective measure is the determination of cyanobacterial presence and quantification through fluorescence.

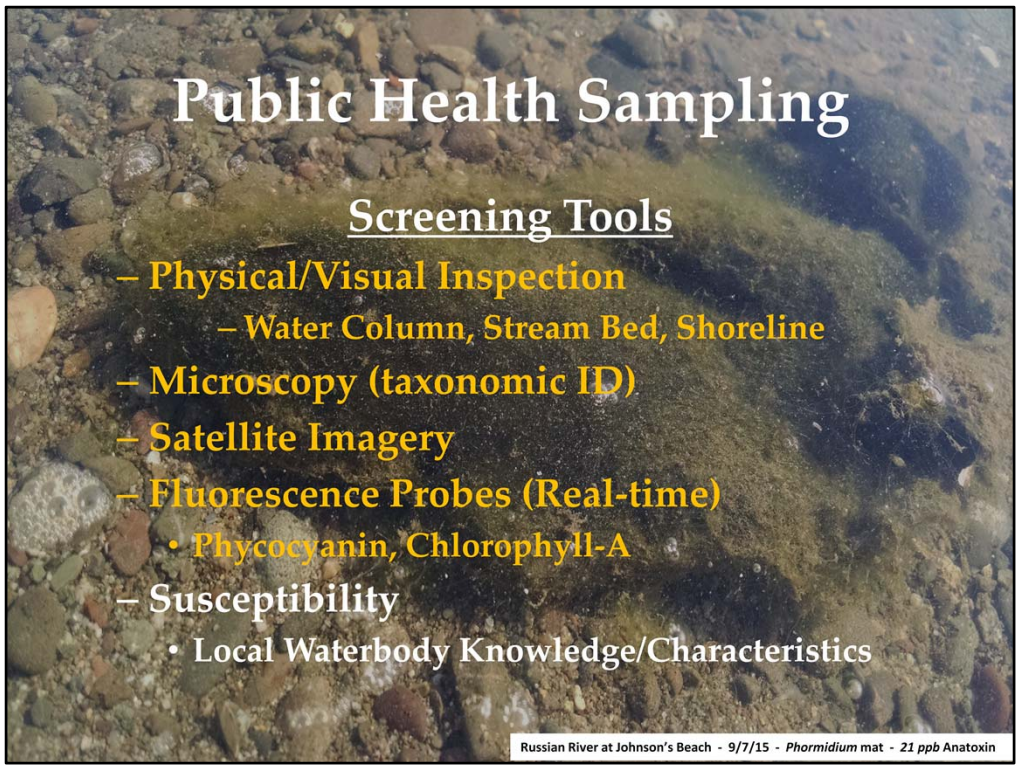
Measurement of Fluorescence

- Chlorophyll-a is common to both cyanobacteria and green algae.
- Phycocyanin is a pigment specific to cyanobacteria.
- Probes can be utilized for in-situ real-time or time series measurements of planktonic cyanobacteria.
- Provides semi-quantitative concentration estimates.

While chlorophyll-A is common to both cyanobacteria and green algae, phycocyanin is specific to cyanobacteria.

Chlorophyll-A and the phycocyanin pigment fluoresce when irradiated with light of a particular wavelength. Because the probes can be calibrated to a standard, the results measured by these probes provide semi-quantitative concentrations of cyanobacteria in the water column. The fluorescence does not provide information on toxin concentrations but rather on relative cyanobacteria biomass.

This tool is predominantly used to determine planktonic cyanobacteria concentration levels and does not capture or represent the benthic cyanobacteria community.



The most effective screening tool that we have available to us is knowledge. Having the knowledge and understanding of the susceptibility of an individual waterbody to cyanobacteria bloom development. Knowing what the local characteristics and triggers are. What are the conditions and/or timing under which those conditions lead to cyanobacteria bloom development.

Public Health Sampling

- **Grab Sample Analysis (Recreational Use Areas)**
  - **Nearshore Water Column**
    - **Shoreline or Nearshore (River / Lake)**
      Shallow Subsurface
    - **Open Water (Lake)**
      Integrated Depth or Shallow Subsurface
  - **Surface Scum**
    - **Shoreline or Nearshore (River / Lake)**
  - **Algal Mats**
    - **Shoreline, Nearshore or Streambed (River / Lake)**

Special Consideration Is For The Most Sensitive Recreators : Children and Pets

So know what?

Once it has been determined that toxin sampling is recommended or necessary, where do you sample for the protection of public health? In rivers, the sample locations are always nearshore or shoreline where young children congregate to play and pets and livestock are most likely to drink. In lakes it can be either nearshore or shoreline or in the open water depending on use. If sampling in the open water, sample timing becomes a critical piece of the sampling protocol as many planktonic species can regulate their buoyancy, moving up in the water column to maximize their ability to photosynthesize and down in the water column to maximize their nutrient intake in stratified waters.

When beginning a monitoring plan, always keep in mind that the most sensitive and susceptible recreators that are most in need of protection are children and pets. For that purpose, public health monitoring should always be focused on nearshore or shoreline areas that are subject to recreational use. Water grab samples should be collected just below the surface in the nearshore recreational areas. Areas with mat and/or scum development should be of particular focus.

Due to the spatial variability of toxin production within a bloom or algal mat, grab sampling of mats and scums should be integrated throughout the recreational area.

The primary purpose of the sampling is to characterize the nature of the bloom in the context of plausible exposure pathways, especially those with potential to harm people and pets. Therefore, samples should target areas where there is the highest likelihood or risk of cyanotoxin interaction and exposure

Toxin Analysis

- **Test Strips**
- **qPCR**
  - **– Quantitative Polymerase Chain Reaction**
- **ELISA**
  - **– Enzyme-Linked Immunosorbent Assay**
- **LCMS**
  - **– Liquid Chromatography–Mass Spectrometry**

Toxin analysis and quantification are generally conducted by a lab. The one exception is the availability of test strips which are generally used in the field, however these are only available .

qPCR does not quantify toxin levels, but is a laboratory tool to isolate and amplify the genetic markers which are present in all cyanobacteria as well as the genetic markers for toxin production. This technique can determine if a sample carries the gene necessary to produce the cyanotoxins microcystin, nodiularian, saxitroxin, and cylindropsermopsin.

ELISA and LCMS analysis provide quantitative results for all the common toxins, including microcystin, nodiularian, saxitroxin, and cylindropsermopsin, as well as anatoxin and lyngbyatoxin.

Ambient (Susceptibility) Sampling

- **Water Quality Risk Monitoring**
  - **Water Quality Risk Factors / Responses**
    - **Nutrients**
    - **Flow Regimes or Lake Levels**
    - **Water Temperature**
    - **Chlorophyll-A**
    - **Phycocyanin**
    - **Dissolved Oxygen**
    - **pH**

Finally...Ambient or Trend monitoring for susceptibility analysis is a monitoring plan to assist us is understanding the characteristics or environmental drivers behind bloom development. This sampling is different then targeted public health monitoring in that we are monitoring for the risk factors behind bloom development.

Elevated nutrient conditions
Increases in water temperature
Decreases in river flows or lowering of lake levels

And evaluating the chemical and biological responses

Increases in Chlorophyll-A and phycocyanin concentrations
And large diurnal swings in pH and dissolved oxygen associated with photosynthesis and respiration

Susceptibility Sampling

- **Public Health Monitoring Data**
- **Physical/Visual Inspection**
  - **Water Column / Stream Bed**
  - **Microscopy (taxonomic ID)**
- **Continuous Data Probes (Time Series Data)**
  - **Phycocyanin and Chlorophyll-A**
  - **DO, pH, Water Temperature**
- **qPCR Monitoring**
- **Satellite Imagery**
- **SPATT Bags**
- **Ambient Nutrient Conditions**

The basic tools in our susceptibility monitoring tool kit are the same as the public health sampling tools, in fact, targeted public health monitoring data and bloom development are the key response parameters.

The difference between the 2 types of sampling are the spatial and temporal components. Susceptibility monitoring is focused on monitoring the risk factors leading up to bloom development so that we can be both predictive and proactive in our public health monitoring as well as determine the causal factors behind bloom development and development management strategies to affect change.

Questions

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