

Efficacy review of a phycocyanin-detecting continuous imaging particle analyzer as used for the identification and enumeration of cyanobacteria (FlowCAM®)

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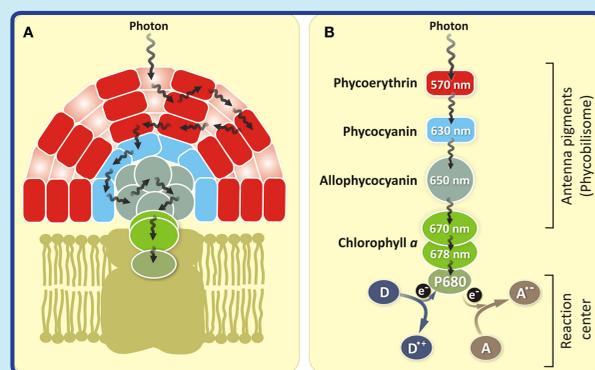


BACKGROUND

Numerous technologies make use of fluorescence measurements to detect and estimate cyanobacteria biovolume in water samples. While useful, results from these instruments can be significantly skewed by turbidity and the presence of other fluorescing pigments and particles.

Fluid Imaging Technologies has recently adapted their imaging particle analyzer, FlowCAM, so that it can detect the phycocyanin pigment in cyanobacteria with the use of an appropriate laser and optical filters. This new instrument can distinguish cyanobacteria from other organisms, relying on pigment detection in addition to image-based morphological analysis.

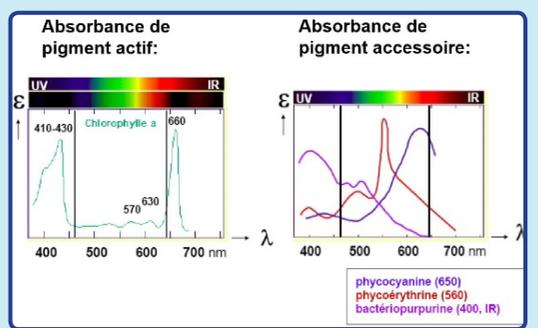
Here we present an overview of this technology and demonstrate its efficacy using cyanobacteria cultures and surface water samples from a public water supply.



Pigments in freshwater cyanobacteria, phycocyanin, act as antenna to direct the photons to the pigments containing the Chlorophyll necessary for photosynthesis.

Front. Plant Sci., 06 July 2011 | doi: 10.3389/fpls.2011.00028
 Adventures with cyanobacteria: a personal perspective
 Govindjee, L. Z. and Dmitry Shevela
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The phycocyanin specifically targets wavelengths not used by Chlorophyll A, therefore thriving in situations where green algae cannot.



Arash Zamyadi

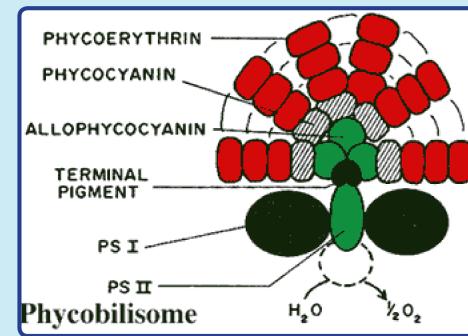
HOW IT WORKS

We selected a laser centered on 630nm to match the natural excitation wavelength of the phycocyanin accessory pigment. To isolate the emission from phycocyanin, we chose two discrete detection filters for our PMT (photomultiplier tubes). One centered on an emission of 655nm and one centered on 700nm wavelength emission. Both are based on peer reviewed literature. The instrument is triggered to take a picture when algae containing phycocyanin are detected.

1. Detect & Capture

The laser in the instrument is directed through a beam shaping filter to create a laser “trip” line.

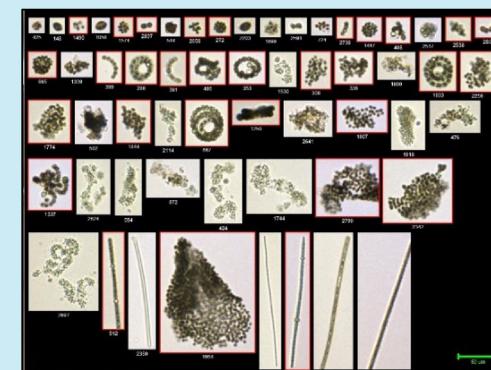
When the algae cross the laser line, IF it contains phycocyanin, the detection filters will pick up the fluorescence and tell the camera to take an image.



http://palaeos.com/eukarya/glossary/glossaryP.htm

2. Tag & Sort

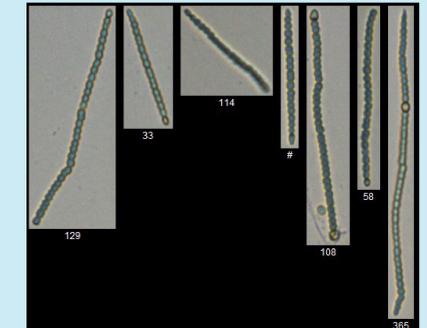
The images taken from the triggering event are uniquely tagged with the fluorescence data from each detection filter. Whether the algae triggered the system on Channel 1 (655) or Channel 2 (700) or both, this info will be available in the post-processing software.



3. Using the Data

Using both the data from the fluorescence detection and standard image parameters, along with the software’s pattern recognition capabilities it is possible to separate images into like types, thereby providing community composition estimates.

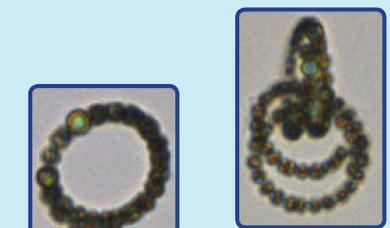
The software provides concentrations, counts, biovolume estimates and size distribution information among other data.



4. Applications

The new FlowCAM for cyanobacteria will support multiple applications.

- Academic Research
- Reservoir Monitoring
- Municipal Water
- Laboratory Research



Anabaena

Anabaena



Microcystis



Microcystis

SUMMARY

With the FlowCAM’s new cyanobacteria detection capabilities, we hope to provide added functionality and time savings to the all users.

We would like to thank scientists at the University of Toronto for their ideas, guidance and assistance in the development and testing of this new technology.