

TECHNICAL BRIEFING

Cell Enumeration of Colonial and Filamentous Cyanobacteria with FlowCam: Method & Case Study

TRACK HAB GROWTH

Accurately determining cell counts of colonial and filamentous cyanobacteria such as *Microcystis* and *Anabaena* is critical for monitoring potentially toxic cyanobacteria. Traditional methods, including manual microscopy, are time consuming, tedious, prone to error and difficult to verify.

In the March 2017 issue of *Harmful Algae*, scientists from the California Department of Water Resources describe how they estimated cell abundance of colonial *Microcystis* using a FlowCam®. This Technical Briefing explains their methodology and also applies it to filamentous cyanobacteria.

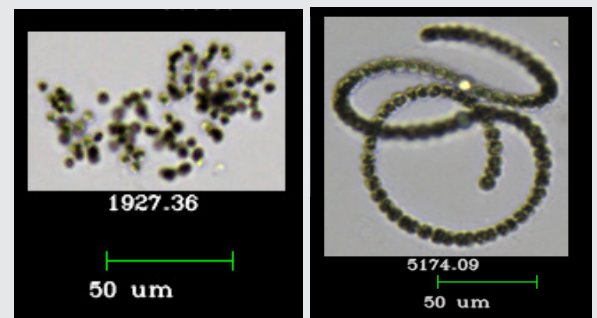


Figure 1. Colonial algae (*Microcystis*) and filamentous algae (*Anabaena*) can be enumerated using the FlowCam and its analysis software VisualSpreadsheet®. Area (ABD) (μm^2) of the colony is noted below each image.

SAMPLE COLLECTION AND PREPARATION

The following excerpts from Lehman et al. (2017), Impacts of the 2014 severe drought on the *Microcystis* bloom in San Francisco Estuary, *Harmful Algae* 63: 94-1208, describe the collection and preparation of their samples.

Net tow samples for determination of Microcystis biovolume (>75 μm size fraction) were preserved with Lugol's solution. The biovolume of Microcystis colonies was computed using Area (ABD) with a FlowCam digital imaging flow cytometer (Fluid Imaging Technologies; Sieracki et al., 1998). In order to more easily measure the biovolume of the colonies, the samples were size fractionated into <300 μm diameter size fractions and read at a magnification of 10X and 4X, and >300 μm and analyzed at a magnification of 2X. Cell abundance estimates based on FlowCam measurements were closely correlated with those determined by microscopic analyses ($r = 0.88$, $p < 0.01$).

Whole water (unpreserved) samples collected from 0.3 m depth were used to determine phytoplankton and cyanobacteria biovolume and taxonomic composition (>10 μm size fraction) in sub-surface water. These samples were kept at 4°C and processed live within 1 to 3 h with a FlowCam digital imaging flow cytometer. The FlowCam was fitted with a fluorescence trigger to isolate live phytoplankton from detritus (Sieracki et al., 1998). Digital images of cells were obtained by passing samples through a 100 mL flow cell for 10 min at 10X magnification.