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Alert levels can be established using baseline data to trigger action in source water management and/or water treatment.

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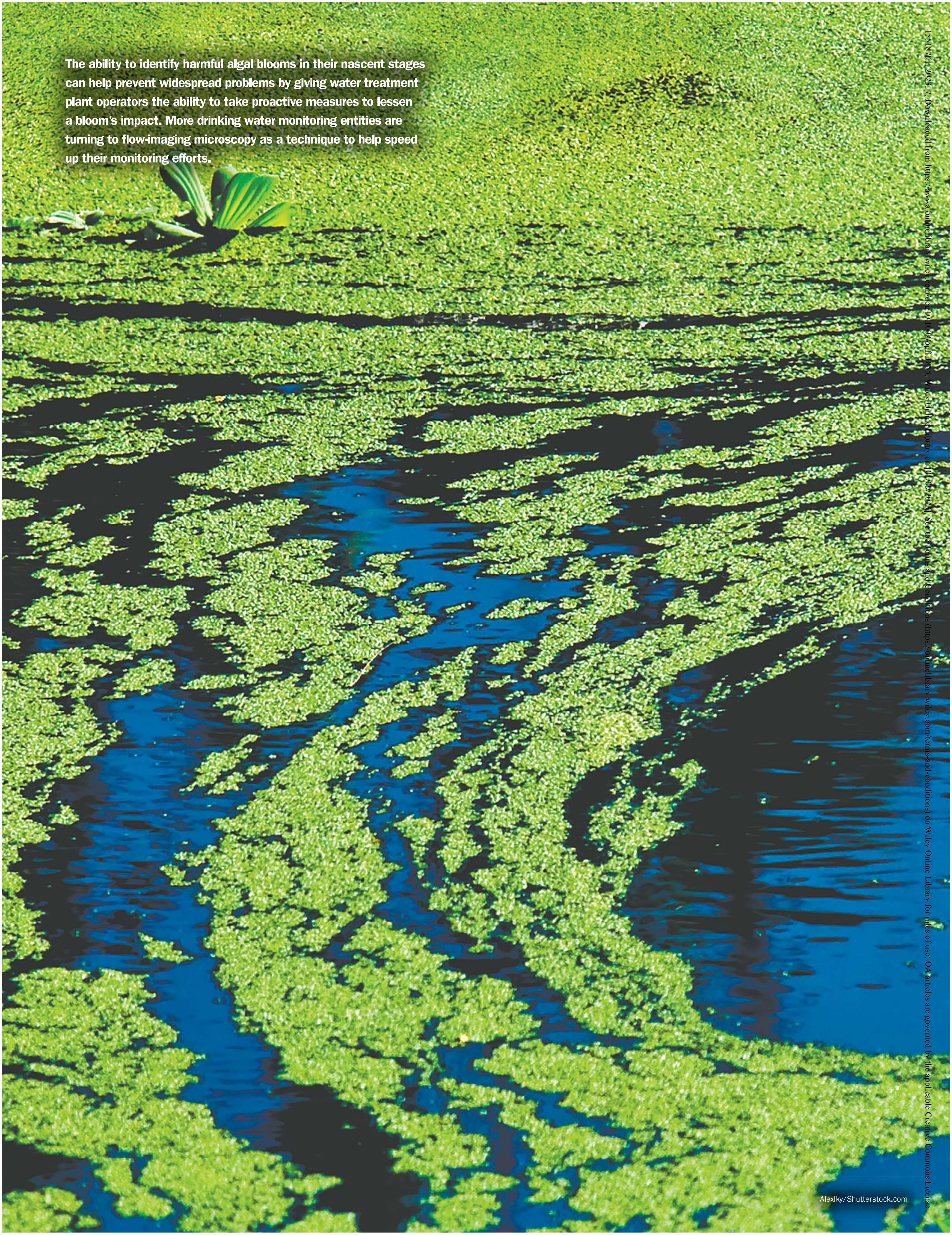
ESTABLISH TRIGGER LEVELS FOR HARMFUL ALGAL BLOOMS

PHYTOPLANKTON, commonly known as algae and cyanobacteria, are defined as organisms that drift with the current. They are at the base of the aquatic food web, sequestering carbon dioxide and generating oxygen through photosynthesis. Variables such as temperature, pH, and nutrient availability can affect the biodiversity of aquatic ecosystems, allowing some organisms to outcompete others. When this happens, predominant organisms can bloom, suppressing the growth of other species and, in some cases, harming the ecosystem balance.

Water bodies undergoing bloom conditions can often become anoxic and experience changes in pH, eventually creating dead zones where aquatic life is no longer able to survive. This is most common in warmer weather when water temperatures are high and dissolved oxygen concentrations are low. In these conditions, there are often noticeable pH swings due to the uptake of dissolved carbon dioxide through photosynthesis as it outpaces cellular respiration.

The organisms responsible for these harmful algal blooms (HABs) in freshwater environments can produce hundreds of compounds known to create taste and odor (T&O) issues in drinking water—the most common being 2-methylisoborneol and geosmin. Some also have genes that code for the production of harmful neuro- and hepatotoxins that can harm humans, livestock, and pets. Recreational water managers are required to monitor for some of these cyanotoxins in certain states, such as Ohio, Oregon, and Pennsylvania, but currently no federal standards exist for phytoplankton monitoring.

The ability to identify harmful algal blooms in their nascent stages can help prevent widespread problems by giving water treatment plant operators the ability to take proactive measures to lessen a bloom's impact. More drinking water monitoring entities are turning to flow-imaging microscopy as a technique to help speed up their monitoring efforts.



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TRADITIONAL VERSUS FLOW-IMAGING MICROSCOPY

The ability to identify blooms in their nascent stages can help prevent widespread problems by giving water treatment plant operators the ability to take proactive measures to lessen a bloom's impact. Integrated monitoring plans provide early warning signs that alert water managers to the need for further field testing. Weekly phytoplankton sampling can confirm these preliminary results and direct further testing. Unfortunately, traditional microscopy methods can be slow, and contracting samples to laboratories for analysis is often pricy, with turnaround time averaging a week or more. Analysis turnaround also depends on the training, taxonomic knowledge, and time spent by the analyst on the samples. In HAB monitoring, time is of the essence, and same-day or next-day results are key to taking prompt treatment action.

As a result, more drinking water monitoring entities are turning to flow-imaging microscopy (FIM) as a technique to help speed up their monitoring efforts. FlowCam from Yokogawa Fluid Imaging Technologies (www.fluidimaging.com) is one example of a benchtop FIM instrument commonly used by drinking water utilities for rapid phytoplankton analysis. Other instruments, such as the Imaging FlowCytobot from McLane Research Laboratories (www.mclanelabs.com), can also be used. When used as part of an integrated monitoring program, FIM can facilitate the development of phytoplankton baseline community compositions as well as HAB trigger or action levels.

Although the need for traditional microscopy and expert taxonomists will never disappear, operators and lake managers generally don't need the specificity gained from a microscope and can use a faster and more reproducible method to classify and enumerate algae and cyanobacteria to the genus level. Users new to FIM can use this article, along with basic information they collect in their source water, to help guide their phytoplankton monitoring programs and adapt to FIM from traditional microscopy.

OVERVIEW AND SAMPLING PROCEDURES

Monitoring Program. A successful HAB monitoring program should answer key questions that allow lake managers to determine the next best course of action, including the following:

- Are there specific monitoring goals in the lake or reservoir?
- What taxonomic groups are predominant in the water system?
- Is this a lakewide event, or is it limited to specific parts of the lake?
- When did the bloom begin, and what is the progression?
- What is the concentration of the predominant organisms?
- What might have caused the bloom, and can any mitigation steps be taken?

Creating an integrated monitoring plan provides a complete picture of lake dynamics. To fully create and understand this picture, it's also important to sample before, during, and after a bloom. Samples taken throughout the year give managers an understanding of the seasonal changes among native populations of phytoplankton (baseline levels). This makes it easier to see shifts in populations as blooms begin to occur and when they reach concentrations of concern or action (trigger levels). Samples taken after a lake has been treated and blooms start to die off demonstrate the effectiveness of the mitigation steps.

The creation of baseline and trigger levels is unique to each utility and lake. There's no one-size-fits-all approach to determine which organisms may become problematic at which levels. However, sampling plans are transferable across water bodies, with the understanding that the organisms and the levels at which they become problematic will differ.

Baseline Levels. Developing a baseline understanding of typical algal diversity and abundance in a lake, reservoir, or coastal area is key to helping users evaluate what is "normal." No two source waters have identical microbiology and chemistry, so it's essential to collect a wide range of water quality data. Baseline levels for this range of parameters, specifically algal diversity, allow users to evaluate future events and assess risk. However, developing a baseline level can be challenging. Phytoplankton populations are constantly changing—seasonally and diurnally as phytoplankton move up and down the water column.

Other factors that might affect natural populations include lake stratification

Table 1. Example Triggers

The City of Wichita Falls Cypress Environmental Laboratory (CEL) set trigger levels low to minimize taste and odor (T&O) issues.

Genus	Trigger Count—*organism/mL*	Type	Reason
Dolichospermum	100*	Cyanobacteria	T&O- and cyanotoxin-producer
Microcystis	150	Cyanobacteria	T&O- and cyanotoxin-producer
Aphanizomenon	150	Cyanobacteria	T&O- and cyanotoxin-producer
Peridinium	400	Dinoflagellate	T&O-producer
Melosira	200	Diatom	T&O-producer
Cyclotella	No limit	Diatom	Abundant but has not been shown to cause an issue in CEL's source water
Pediastrum	No limit	Green algae	Abundant but has not been shown to cause an issue in CEL's source water

*chains/mL

Flow-imaging microscopy is a powerful part of any source water manager's toolkit and can be used to make data-driven treatment decisions in source water and treatment plants.

and nutrient availability. Aquatic ecosystems are extremely dynamic, and it's important to consider that it may take multiple seasons to fully understand the changes in algae populations in a water body and that these populations tend to be unique to that location. When planning a new monitoring program, at least one season of monitoring—ideally up to two or three—should be part of the creation of these baseline levels.

Trigger Levels. Trigger levels can be defined as the point at which the concentration of one genus of phytoplankton reaches a number that prompts further sampling and/or treatment. These levels aren't universal and should be developed from monitoring baseline populations and concentrations. These numbers should be reviewed each year to determine whether updates are needed. By continually evaluating the program, baseline data sets grow and help managers make informed decisions.

Figure 1. Comparison of Two Flow-imaging Microscopy (FIM) Instruments

Tulsa purchased an upgraded FIM system capable of laser-based pigment differentiation of cyanobacteria versus other algae and diatoms.

RESULTS IN ACTION

Consider the HAB monitoring plans for three drinking water utilities and the steps these utilities took to develop their baseline and trigger levels. Each of the following utilities uses FIM to quickly analyze data from multiple samples on a set schedule. These case studies are only examples; the trigger levels provided here shouldn't be used without evaluating them in each user's region.

City of Wichita Falls, Texas. The City of Wichita Falls Cypress Environmental Laboratory (CEL) implemented FIM for HAB monitoring in 2016. Understanding that the transition from traditional microscopy to FIM wouldn't be a directly proportional change, staff committed to establishing an in-depth baseline. Samples were collected and analyzed five days per week for one full year. This benefited CEL by finding that

- seasonal variation is a main driver for algal population dynamics.
- small blooms producing T&O were observed even in cooler months.
- each reservoir has unique algal populations, and types (green algae, diatoms, dinoflagellates, cyanobacteria) often follow predictable bloom cycles.
- time of day, sampling location, and sampling depth can directly affect organism counts.
- rain runoff often triggers blooms.
- cyanobacteria are the main nuisance organism for the city's region.

After the baseline year was complete, analysis was scaled back to seasonal, with samples analyzed during warmer months (June–August) three times per week, winter months (December–February) once per week, and spring/fall (March–May and September–November) twice per week. These schedules are kept unless spikes in particular genera are seen. Example triggers can be seen in Table 1. Triggers set after completing the baseline year were also much lower than the previous trigger levels assigned to the data supplied via traditional microscopy.

Although these triggers are part of CEL's monitoring plan, they are always subject to adjustment based on new findings. If a genus is seen to cause an issue at a lower organism count, triggers are reduced to continue providing early warning for the system. For organisms such as *Cyclotella* and *Pediastrum* that haven't been problematic for CWF, they don't even need to be counted. Software collects the data that can be processed at a later date if needed. A proactive monitoring program is continually reevaluated to maintain its effectiveness, and triggers should be established at limits lower than the actual count seen to cause issues so the system has time for an early response.

City of Tulsa, Okla. The Water Quality Assurance laboratories for the City of Tulsa first began using FIM for phytoplankton monitoring in 2008, replacing manual microscopy. Following a year of baseline monitoring of the city's two watersheds using this system, and in conjunction with a literature review of current published plans, an early warning plan (EWP) was developed. It was determined that in these watersheds, it takes about a year to have a clear understanding of the typical phytoplankton seasonal succession. The

Table 2. Early Warning and Treatment Consideration Levels

Massachusetts Water Resources Authority increases monitoring if early warning levels are exceeded for algal abundance.

Genus	Early Warning Trigger Count—*ASU/mL*	Treatment Consideration Trigger Count—*ASU/mL*	Type	Reason
Dolichospermum	>15	>25	Cyanobacteria	T&O- and cyanotoxin-producer
Chrysosphaerella	>100	>500	Chrysophyte	T&O-producer
Synura	>10	>12	Chrysophyte	T&O-producer
Dinobryon	>200	>500	Chrysophyte	T&O-producer
Uroglenopsis	>200	>750	Chrysophyte	T&O-producer

ASU—areal standard unit, T&O—taste and odor

EWP clearly outlined contingency steps that would be put into action once cyanobacteria populations reached certain trigger levels and addressed next steps for T&O issues stemming from cyanobacteria blooms, cyanotoxin contingency plans, and treatment plans for the reservoirs and water treatment plants.

In 2021, the City of Tulsa purchased an upgraded FIM system capable of laser-based pigment differentiation of cyanobacteria versus other algae and diatoms. Concurrent data were collected from the new instrument to compare with the data routinely gathered using the previous system (Figure 1). Samples were run on both systems for a year to compare data.

On first inspection, there was no apparent correlation between the two data sets. Factors affecting the differences included improved electronics, a field-of-view flow cell, and a higher-resolution camera, each of which contributed to the increased and more accurate results. After further analysis, the results were converted to log scale, at which point a trend emerged.

As part of the method development, the laboratory purchased a pure culture of *Dolichospermum circinalis* and compared the particles-per-milliliter count, using FIM, with the natural unit count, using traditional microscopy. Results showed that the natural unit count from the microscope was higher but trended the same as FIM particles per milliliter. This offered assurance that the values obtained using FIM are a reasonable representation of a sample.

Massachusetts Water Resources Authority. Massachusetts Water Resources Authority (MWRA) monitors for T&O algae/cyanobacteria in its two source reservoirs, the Quabbin and Wachusett. Algae-related T&O complaints were a routine seasonal problem through the 1980s and 1990s. Algae trigger-level data were first implemented in late 1997 from 1990–1996 data. These levels have been updated as more data have become available. MWRA began using FIM in June 2004. FIM samples are collected in the Wachusett Reservoir by MWRA once per week from May through September. Traditional microscopy is used by the Massachusetts Department of Conservation and Recreation to monitor algae/cyanobacteria in the Wachusett Reservoir weekly and in the Quabbin Reservoir once every two weeks.

MWRA uses areal standard units (ASU)/mL to measure algal abundance. To help compare data with other utilities that use the unit of measure of cells per milliliter, MWRA used an estimate of 1 ASU/mL as roughly equal to 100 cells/mL. However, the actual conversion of ASU/mL to cells/mL depends on the type and size of algae. Still, this provided context for understanding data using different measurement units. If early warning levels are exceeded (Table 2), monitoring is increased, and compilation of customer complaint data occurs more frequently. If treatment consideration levels are exceeded, a review of in-reservoir and in-water treatment plant options will occur. In addition, regulatory agencies and the public will be notified.

Algae count data, coupled with customer complaint data, have driven the current trigger level values. Since ozone treatment was added to the Carroll Water Treatment Plant, MWRA has received fewer complaint calls about T&O. *Chrysosphaerella* levels have exceeded 1,000 ASU/mL, with no customer complaints. As a result of this added treatment, MWRA is working to update these levels to balance the needs of the consumer and concerns of over-treatment of the reservoir with copper sulfate. Although T&O complaints have been well controlled, concerns about cyanobacteria and cyanotoxins must be considered in response plans. Additional water quality data, such as chlorophyll-a, 2-methylisoborneol/geosmin, the ratio of dissolved inorganic nitrogen to phosphorus, UV_{254} (ultraviolet absorbance at 254 nm), and chlorine demand/decay, are being evaluated as indicators of T&O or cyanobacterial algae blooms.

AN EFFECTIVE TOOL

Although there's no magic button to automatically produce a customized workflow and triggers, FIM is a powerful part of any source water manager's toolkit and can be used to make data-driven treatment decisions in source water and treatment plants. Long-term baseline data allow users to establish effective triggers, which should be continually monitored and adjusted as new data are collected. FIM provides a higher throughput analysis than manual microscopy, and triggers can be tailored to genus-level identifications. When used proactively, FIM can save users time and money—both in the laboratory and in water treatment.