Report

Report on

Cyanobacteria: Initial Assessment of New England Water Supplies

Comprehensive Environmental, Inc.

In association with
The University of New Hampshire
Center for Freshwater Biology

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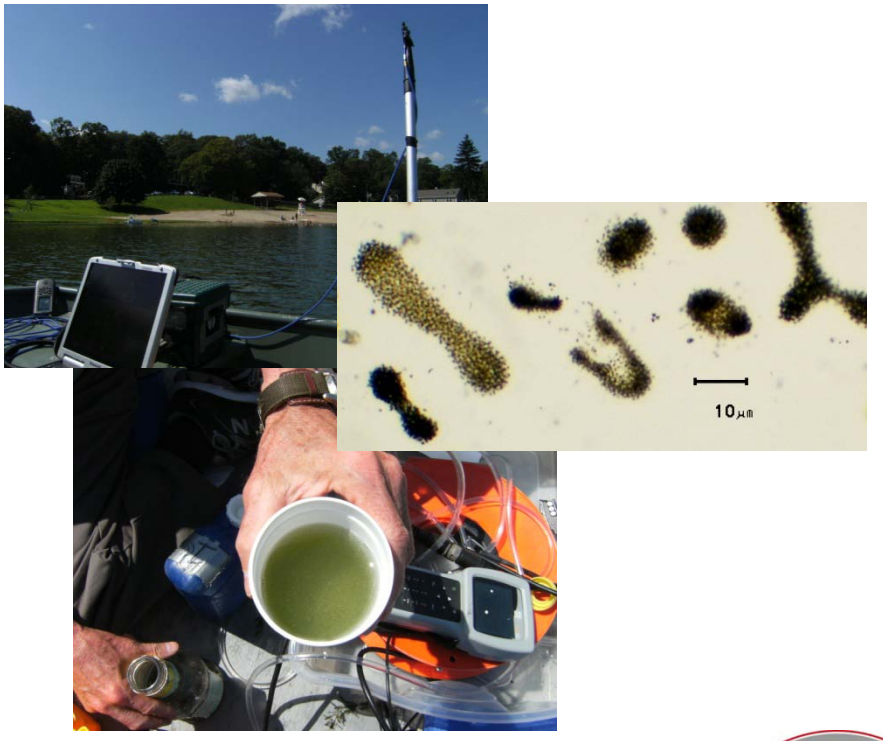

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Executive Summary

Alarming levels of potentially toxic cyanobacteria have occurred in some New England water bodies, including drinking water sources, in recent years. Cyanobacteria blooms have caused serious problems in water bodies, such as fish kills and pet deaths. Also, a recent study identified possible links between high levels of cyanobacteria in recreational lakes and ALS (Lou Gherig's) type diseases in New Hampshire lakes.[1] These events have caused some degree of public alarm at the time they occurred, and more questions and/or concerns may be expected if water supplies are involved.

Exposure to cyanobacteria could be through drinking water, aerosols or skin contact. Recent advances in the ability to detect lower levels of cyanotoxins and epidemiological studies examining cyanotoxin effects on human health have heightened concerns. Cyanotoxins can cause a range of human health issues such as liver and kidney damage, neurological damage, gastrointestinal issues, and tissue damage. The risks for drinking water supplies is not well known, but likely depends on the treatment process as well as how "slugs" or mats of the cyanobacteria are handled when they enter the treatment facility.

Although some water systems are all too familiar with the challenges associated with taste and odor issues caused by some cyanobacteria, concerns about human health effects are more recently coming to light. In an effort to determine the magnitude of the cyanobacteria and cyanotoxin presence in New England drinking water supplies, Comprehensive Environmental Inc. (CEI) conducted an initial assessment on cyanobacteria and microcystins removal at four New England water treatment facilities. CEI is a progressive civil and environmental engineering consulting firm, striving to stay ahead of issues affecting our industry. Through these efforts, we provide our clients with the highest level of service and potentially pass on new information to the drinking water community. For this initial assessment, CEI collaborated with the University of New Hampshire, Center for Freshwater Biology and four New England drinking water systems to determine (for the first time) whether cyanobacteria and microcystins (liver toxins produced by many species of cyanobacteria commonly found in New England) are effectively removed through water treatment processes.

[1] Stommel, Dr. Elijah, Dartmouth-Hitchcock Medical Center. January 10, 2010. *Possible Links between Cyanobacterial Blooms and ALS*. Presentation at the New England Interstate Water Pollution Control Center's Chelmsford workshop on Cyanobacteria.

The results of the initial assessment indicate that the amount of microcystins (one type of cyanotoxin) in the drinking water supplies tested were well below the World Health Organizations recommended guideline of 1,000 nanogram per liter (ng/L). Select water treatment processes were able to reduce the amount of microcystins in the water. However, there were no cyanobacteria blooms and levels were fairly low entering the treatment processes during this limited study; it is not known how the treatment processes would respond to a cyanobacteria bloom entering as a slug. Further research is needed in order to determine how well various treatment processes will remove larger concentrations of cyanobacteria.

Section 1.0
Cyanobacteria

1.1 Cyanobacteria Overview

Cyanobacteria, formerly known as blue-green algae, are photosynthetic organisms that often occur in fresh, brackish or marine waters, thriving particularly in nutrient rich, warm waters. They are generally more competitive than algae and may survive in sediments from year to year. Cyanobacteria grow as unicellular, colonial and filamentous forms and may produce pigments other than blue-green including black, olive, and red. When cyanobacteria concentrations increase, they often form visible blooms, surface scums or benthic mats. In addition to aesthetic color issue, these large populations can produce compounds that cause taste and odor issues such as geosmin and methyl isoborneol (MIB). More recently concerns have increased regarding the toxic compounds cyanobacteria are capable of producing, harmful to humans and other animals, referred to as cyanotoxins (AWWA, 2010).

Cyanobacteria are widespread and have been studied for many years, however, recent research and documentation of cyanobacteria and their toxic effects has highlighted increased concerns about these organisms and their potential health and environmental effects. Since temperature and nutrients seem to be a driving force for growth, the increasing water temperatures occurring with climate change and higher levels of stormwater runoff from increasing urbanization of water supply watersheds may exacerbate the problem.

The United States Environmental Protection Agency (USEPA) now encourages awareness of cyanobacteria in drinking water, and has funded studies examining cyanobacteria and associated health effects caused by cyanotoxins. As a result, the USEPA currently lists three cyanotoxins on the Safe Drinking Water Act's Contaminant Candidate List (CCL3) and Unregulated Contaminant Monitoring Rules (Anatoxin-a, Microcystin-LR, and Cylindrospermopsin).

At the state and local levels, authorities in all New England states are now monitoring for cyanobacteria and have closed recreational use of some lakes and ponds when blooms are detected (NEIWPCC Regional Cyanobacteria Workshop, Chelmsford, MA, January 13,

2010). While drinking water maximum contaminant levels (MCLs) for cyanobacteria and cyanotoxins have not been established by the USEPA, the World Health Organization (WHO) established a guideline of 1.0 microgram per liter (μg/L) or 1,000 nanogram per liter (ng/L) for microcystin-LR, one type of cyanotoxin (WHO, 1999). However, this does not consider exposure to the other types of cyanotoxins so some researchers suggest that this level is too high.[2]

1.2 Types of Cyanobacteria

There are different species of cyanobacteria, each suited to grow in different environments. Common fresh water cyanobacteria include: *Anabaena, Aphanizonmenon, Cylindrospermopsis, Lyngbya, Microcystis, Planktothrix, Woronichinia, Anabaenopsis, Nostoc, and Phormidium*. Since they require light for growth, cyanobacteria are more likely to be found in surface waters, but they also may be transported into groundwater.

The following cyanobacteria genera known to produce geosmin also produce cyanotoxins: *Anabaena, Aphanizonmenon, Lyngbya, Microcystis, Oscillatoria,* and *Phormidium* (WHO, 1999). At this time, however, no reliable correlation has been found between odor levels and toxin production. Taste and odor issues may indicate the presence of cyanotoxin producing cyanobacteria, however, this should not be the only method used for detection of cyanobacteria. Studies show that the human nose can detect geosmin at concentrations as low as 5 parts per trillion and that taste buds can detect geosmin at 0.7 parts per billion (USEPA, 2009).

Cyanobacteria produce numerous types of cyanotoxins. The cyanotoxins are produced and contained within growing cyanobacteria cells. Generally, release of cyanotoxins occurs during cell death and lysis, however, some types of cyanobacteria release cyanotoxins during growth if light conditions are poor. Research into the frequency and effects of these toxins is ongoing. However, it is generally thought that microcystin-LR is the most frequent and probably most toxic of the microcystins.

[2] Haney, Dr. James. Center for Freshwater Biology, University of New Hampshire, December, 2010. Personal Communication.

1.3 Toxins and Effects of Toxins

There are three known classes of cyanotoxins including cyclic peptides, alkaloids and endotoxins (AWWA, 2010). Cyclic peptides include hepatotoxins. Alkaloids include neurotoxins, cytotoxins and dermatotoxins. Endotoxins include lipopolysaccharides. Cyanobacteria commonly found in surface water supplies produce cyanotoxins including hepatotoxins and neurotoxins. Cyanotoxins can cause a range of symptoms depending on the type of toxins and concentration. Symptoms include stomach cramps, vomiting, diarrhea, fever, headache, pains in muscles and joints, skin, eye and throat irritation, kidney dysfunction, liver damage, and death.

Hepatotoxins are produced by several cyanobacteria species. Most hepatotoxins are microcystins. There are numerous types of microcystins that may be produced during a bloom. These microcystins have different levels and types of lipophilicities and polarities that affect toxicity. Microcystin-LR has been studied the most, since it is the most frequent and most toxic microcystin. Microcystin-LR is produced by many species including *Anabaena, Limnothrix, Microcystis, Oscillatoria* and *Planktothrix* (AWW, 2010). Microcystins will degrade naturally in water and generally have a half life of less than one week (WHO, 2003). Acute exposure to microcystins causes severe liver damage and can lead to heart failure and death. Chronic exposure can cause liver damage as well.

The alkaloids are not found as widespread in water supplies. Neurotoxins are highly toxic nerve poisons with short half-lives, so they do not have chronic exposure affects like microcystins. Ingestion of neurotoxins can cause death depending on the amount and species ingested (WHO, 2003). Cytotoxins affect the liver and kidneys. Dermatotoxins affect the skin causing rashes (AWWA, 2010).

Endotoxins can also be found in cyanobacteria. These include lipopolysaccharides that can cause gastrointestinal distress (AWWA, 2010).

Table 1-1 provides a brief summary of the groups of cyanotoxins.

Table 1-1
Groups of Cyanotoxins

Group	Select Sub-Groups	Cyanotoxins on USEPA CCL3	Human Health Affects
Cyclic Peptides			
	Hepatotoxins		Liver Damage, possible carcinogen
		Microcystins	
Alkaloids			
	Neurotoxins		Neurological Damage
		Anatoxin–a	
	Cytotoxins		Liver and Kidney Damage
		Cylindrospermopsin	
	Dermatotoxins		Skin Rashes
		--	
Endotoxins			
	Lipopolysaccharides		Gastrointestinal Distress and tissue irritant
		--	

Three cyanotoxins are on the USEPA CCL3: hepatotoxin microcystin, neurotoxin anatoxin–a, and cytotoxin cylindrospermopsin. These toxins were listed as priority contaminants in 2001. The CCL3 list was proposed in 2009 and the regulatory determination is anticipated for 2013. The USEPA is currently evaluating the effects of these cyanotoxins on human health. While it is unlikely that treated drinking water will contain high concentrations of cyanobacteria and cyanotoxins, more research is needed as to the risks associated with low-level long-term exposure.

1.4 Environmental Fate of Cyanobacteria and Toxins

Cyanobacteria grow and flourish in water environments rich in inorganic nutrients including nitrogen and phosphorus. Phosphorus in particular appears influential in the explosive growth of these organisms. Other factors such as water temperature and pH also affect growth. Optimum temperatures are between 15 and 30^{o}C and optimum pH between 6 and 9 pH units (WHO, 2003). Generally, blooms occur in late summer and fall and become more established when conditions are calm and may persist for 2 to 4 months. Some blooms may survive underneath the ice in surface water throughout the winter creating a year round problem.

The cyanobacteria species require different amounts of daylight with some adjusting buoyancy in response to changing available light. Light and temperature affect the release of toxins. Additionally, a larger amount of toxin will be released when the bloom dies all at once, as compared to the amount of toxins released by a growing population of similar size. The levels of toxins within water bodies will increase significantly days after application of algaecides (i.e., copper sulfate), before degrading after several weeks.

Table 1-2 presents the major parameters that influence cyanobacteria growth, specifically *Microcystis* and *Anabaena*. This table may be used as a general guide.

Table 1-2
Major Parameters Influencing Cyanobacteria Growth
(Source: *International Guidance Manual for the Management of Toxic Cyanobacteria,* Global Water Research Coalition)

Potential for Cyanobacteria Growth	History of Cyanobacteria	Water Temperature (^{o}C)	Nutrients Total Phosphorus (µg/L)	Thermal Stratification
Very Low	No	<15	<10	Rare or Never
Low	Yes	<15-20	<10	Infrequent
Moderate	Yes	20-25	10-25	Occasional
High	Yes	>25	25-100	Frequent and Persistent
Very High	Yes	>25	>100	Frequent and Persistent/strong

Section 2.0
Cyanobacteria and Water Treatment

2.1 Impacts of Cyanobacteria on Drinking Water

Cyanobacteria and cyanotoxins are a concern with regards to providing aesthetically pleasing and safe drinking water, in addition to recreational resources. Cyanobacteria can affect drinking water by producing compounds causing earthy and musty odors (geosmin and MIB). As described in Section 1.0, cyanotoxins can cause adverse health effects when ingested by humans including gastrointestinal issues, liver damage and even death, if consumed in great quantity.

The WHO has established a drinking water guideline of 1.0 μg/L for microcystin-LR, a type of cyanotoxin and one of more than 80 variants of microcystin. Additional research is ongoing to determine appropriate guidelines for other cyanotoxins. The USEPA is currently evaluating the appropriate guidelines and/or regulations for cyanobacteria and cyanotoxins accounting for the human health risks and available drinking water treatment practices. A regulatory determination is anticipated to be made for three cyanotoxins in 2013 including hepatotoxin microcystin, neurotoxin anatoxin–a, and cytotoxin cylindrospermopsin. It is highly likely that the WHO guideline is too high for drinking water in the U.S.A. as it does not consider any cumulative effects of other cyanotoxins.

Protection against intracellular and extracellular cyanotoxins may be achieved through water resource protection and/or removal by treatment. The strategies used will depend on which cyanotoxins are present, how much cyanotoxin is in the raw water supply, the required finished water concentration and the abilities of treatment strategies to remove cyanotoxins.

2.2 Source Water Protection and Control

Cyanobacteria growth is encouraged in waters high in nutrients such as nitrogen and phosphorus. Additional factors affecting growth of cyanobacteria include temperature, light, micronutrients (iron, molybdenum), pH, alkalinity, buoyancy, hydrologic and meteorologic conditions, and the morphology of the impoundment (Hitzfeld, 2000).

2.2.1 Source Water Protection

Limitation of nutrient and sediment loading will help limit the potentially explosive and dangerous growth of cyanobacteria blooms. Nitrogen and phosphorus enter water bodies from surface runoff and erosion, stormwater discharge and wastewater discharge. In some cases, sediment buildup and anoxic conditions may also lead to internal phosphorus cycling that can result in even greater phosphorus loads. Controlling land uses, maintaining landscape integrity and best management practices can be employed to help limit cyanobacteria growth. Comprehensive watershed management programs may include (among others) control of stormwater and other pollutant inputs of phosphorus from suburban/urban lands and construction sites; waterfowl controls; and in-lake controls described below.

2.2.2 Source Water Cyanobacteria Control

Source water cyanobacteria control includes measures such as nutrient precipitation, dredging, use of algaecides and use of ultrasonic sound wave equipment.

Phosphorus levels available for uptake by cyanobacteria may be reduced by changing dissolved phosphorus into a precipitate so that it gets bound in the sediment. The phosphorus load to the water body must be low and the water must be deep so that the material is not re-suspended. Several types of chemicals have been used for phosphorus precipitation including aluminum sulfate, ferric salts, and lime. Use of these chemicals must be approved by the state. The addition of chemicals is not commonly practiced with natural waters in the present day due to increased awareness of the effects of these chemicals on aquatic life and the localized ecosystem. Aeration has also been used to reduce the stratification that usually produces anoxic conditions in lakes and reservoirs.

Algaecides, such as copper sulfate, have historically been used to reduce algae blooms in water bodies. However, it has been shown that treating a surface water with copper sulfate after a bloom may exacerbate the cyanotoxin issues. Copper sulfate kills the cyanobacteria releasing the intracellular cyanotoxins into the water. This makes the cyanotoxins more difficult to remove by conventional treatment. Copper sulfate is only recommended as a last resort due to the cyanotoxin release and the addition of

copper to the water body (WHO, 1999). Most states require permits for the use of algaecides.

New technologies for algae control without the use of chemicals are emerging, for example, there is now equipment that produces ultrasonic sound waves that may eliminate some algae and presumably cyanobacteria by interrupting the life cycle of the cells. Ultrasonic equipment requires line of sight devices, so for more irregular shaped waters more equipment is needed. These devices can help to reduce algae within several days to weeks, but do not affect the source of the problem and can be expensive to operate.

2.3 Treatment Removal Methods

The ideal treatment for cyanobacteria would be removal of cyanobacteria without causing toxin release. Once toxins are released treatment processes need to be capable of removing the soluble toxins. There have been studies on the removal capability of various treatment processes. Some of these processes are effective at removing the cyanobacteria, but not the extracellular cyanotoxins. A selection of the typical treatment processes are described below.

2.3.1 Coagulation and Clarification

Coagulation and clarification have been shown to be effective processes for cyanobacteria and algae removal. The chemical type, dosages and water quality greatly affects the optimization of cyanobacteria removal. Cyanobacteria are the last phytoplankton cell to be removed at insufficient coagulant dosages (WHO, 1999). The optimum chemical dosages may be determined by measuring the zeta potential (electrophoretic mobility) of cells. Studies have shown that the coagulant dosage needed is proportional to the sum of the alkalinity and logarithm of the cell number (WHO, 1999). Additionally, it has been shown that smooth, spherical cells coagulate by charge neutralization, while filamentous algae or species with bristles require sweep coagulation (WHO, 1999). Coagulation is an effective method for removal of intact cyanobacteria cells, but not toxins already separated from the cells.

Once the cyanobacteria are trapped within the clarification residuals, the toxins will be released within days of treatment. Residuals collected from the clarification process must be regularly removed from the clarifier to avoid release of cyanotoxins into the clarified water. If the treatment facility includes recycle water from residuals handling basins, this may increase the amount of cyanotoxins in the treatment stream. Coagulation/clarification is not considered an effective means of treatment of extracellular cyanotoxins (Westrick, 2008).

2.3.2 Filtration

Rapid filtration alone is not sufficient for removal of cyanobacteria. Conventional treatment is needed (ie. coagulation/clarification/filtration) for effective removal of cyanobacteria and intracellular cyanotoxins. Studies show that little cell breakage occurs during filtration (Westrick, 2008). The filters need to be backwashed regularly so as to remove the cyanobacteria before the dead cells release cyanotoxins into the filter.

Slow sand filtration allows for the formed filter cake and biofilm to perform most of the filtration. Studies have shown that up to 99% of algal cells are removed by slow sand filtration (WHO, 1999).

Absorptive filters including powdered activated carbon (PAC) and granular activated carbon (GAC) have been shown to provide successful removal of cyanobacteria and microcystins. However, high dosages of PAC are needed to attain high removal rates of microcystins. Studies have shown that GAC is effective in removal of microcystins, as long as the adsorption capacity of the GAC is not exhausted. Removal using GAC is also dependent on the degree of biological activity on the GAC (high EBCT has been shown to help removals) and the magnitude and duration of the occurrence (WHO, 1999).

Membrane treatments using microfiltration (MF) and ultrafiltration (UF) may be effective in removal of cyanobacteria and microcystins. However, since MF has a larger pore size it may not be effective in removing all intact cyanobacteria and cyanotoxins; additionally, UF cannot removal all of the extracellular cyanotoxins

(Westrick, 2008). Therefore, membrane treatment is effective in removing some cyanobacteria and intracellular cyanotoxins, but must be used in combination with other treatment processes. Reverse osmosis (RO) or nanofiltration can remove cyanobacteria, intracellular and some extracellular cyanotoxins (AWWA, 2010).

2.3.3 Oxidation and Disinfection

Studies have shown that cyanobacteria microcystins can be degraded by strong oxidants such as chlorine, ozone and potassium permanganate. Potassium permanganate has been shown to be successful in removal of microcystin-LR and anatoxin-a (AWWA, 2010).

Ozone has been shown to successfully destroy microcystins, but only after the Dissolved Organic Carbon (DOC) demand is satisfied (WHO, 1999). It is also believed that ozone is effective in inactivating cylindrospermopsin and anatoxin-a, but not saxitoxin, however, dissolved carbon will compete with destruction of cyanotoxins and their destruction is effected by alkalinity and temperature (Westrick, 2008). Ozone may be added as pre or post treatment. Increases in cyanobacteria will increase the ozone demand. If cyanobacteria and organic levels in the raw water increase unexpectedly and the ozone demand is not increased, incomplete degradation of cyanotoxins will occur (Hitzfeld, 2000).

Studies show that chlorination is an effective means for destroying microcystins. The addition of chlorine for disinfection can help to degrade microcystins, provided there is a free chlorine residual of 0.5 mg/L after 30 minutes contact time with pH less than 8.0 pH units (WHO, 1999). Additionally, chlorination can be used to inactivate cylindrospermopsin and microcystins at lower pH values (pH 6-7) and saxitoxins at higher pH values (pH 9), while anatoxin-a may not be degraded by chlorination. Chloramines and chlorine dioxide do not appear to appreciably degrade cyanotoxins (Westrick, 2008).

Surface water treatment facilities generally already use disinfection for inactivation of viruses (4-log removal), *Giardia* (3-log removal) and *Cryptosporidium* (2-log removal). The amount of disinfection or removal needed using chlorination depends on the water pH and temperature, amount of chlorine and contact time provided and the amount of log removal provided by other treatment processes at the facility. Assuming a pH range of 6.0 to 9.0 and a water temperature of 10^{o}C (50^{o}F), a CT-value of 6.0 mg/L-min is needed to achieve 4-log removal. With a chlorine residual of 0.5 mg/L, this equates to a contact time of 12 minutes. This means that if 30 minutes of contact time are needed with a chlorine residual of 0.5 mg/L for microcystin degradation, it is unlikely that most water treatment facilities provide full microcystin degradation through chlorination alone.

The doses of UV needed for microcystin, cylindrospermopsin, and anatoxin-a destruction are several orders of magnitude higher than that needed for *Cryptosporidium* oocyst inactivation, making it not economically feasible for cyanotoxin inactivation (Westrick, 2008).

2.3.4 Summary

Removal and inactivation of cyanobacteria and intracellular and extracellular cyanotoxins requires a combination of treatment processes or a multiple barrier approach. Each treatment facility is unique and may need to be examined specifically for cyanobacteria and cyanotoxins. Intracellular cyanotoxins may be removed by conventional treatment and membrane filtration. Extracellular cyanotoxins may be removed and/or degraded by chlorine at varying pH values and longer contact times, or by potassium permanganate, ozonation, carbon adsorption and reverse osmosis. At this time it does not appear that chloramination, chlorine dioxide, hydrogen peroxide or UV treatment are effective means of extracellular cyanotoxin inactivation (Westrick, 2008).

Water systems using source waters that contain or may contain moderate or high levels of cyanobacteria and cyanotoxins (surface waters and some groundwaters) should consider development of a source to tap management program. Water samples can be tested for cyanobacteria and cyanotoxins in the field or by laboratories as

discussed in Section 4.0 of this report. However, this grab sampling method is subject to the presence of cyanobacteria and/or cyanotoxins in the water at the time of sampling. Cyanobacteria move around in water bodies vertically with changes in stratification and light and can move across the horizontal plane by wind and currents. Therefore, there is a high potential of missing water containing cyanobacteria and/or cyanotoxins using the grab sample technique. In-line monitoring equipment is available for continuous monitoring of cyanobacteria. Phycocyanin blue-green algae sensors can be installed to monitor for cyanobacteria. These sensors measure the fluorescence of phycocyanin in the living cyanobacteria cells. In-line monitoring can provide operators with an early warning system for increasing cyanobacteria biomass that may indicate a potential taste and odor event and/or cyanotoxin increase.

Section 3.0
Site Specific Cyanobacteria Testing

3.1 Test Locations

Comprehensive Environmental Inc. conducted an evaluation of the ability of four New England water treatment facilities to remove cyanotoxins from the raw water supply. Samples from the raw water, finished water and at key points in the treatment trains (ie. filtered water, etc.) were collected for analysis during the summer of 2010.

3.1.1 Water System 1

Water System 1 is a surface water treatment facility utilizing super pulsator clarification, filtration and chlorine disinfection. Raw water consists of a combination of reservoir surface water and groundwater. Water can be withdrawn from the 22 feet deep reservoir at several different depths, but the typically selected depth to obtain optimum raw water quality is 5 to 10 feet below the average water surface. Weirs and floating baffles are used to prevent algae growth. Facility chemical feed includes ferric chloride (coagulation), sodium hydroxide (pH adjustment), non-ionic polymer (coagulation aid), sodium hypochlorite (disinfection) and zinc orthophosphate (corrosion control).

3.1.2 Water System 2

Water System 2 is a conventional surface water treatment facility utilizing flocculation, sedimentation (basin), rapid dual media filtration including powdered activated carbon (PAC) and chlorine disinfection. Raw water is obtained from a river. Settled residuals are removed daily and filters are backwashed about every other day. Facility chemical feed includes soda ash (pH and alkalinity adjustment), polyaluminum chloride (coagulation), sodium bicarbonate (pH adjustment), and sodium hypochlorite (disinfection).

3.1.3 Water System 3

Water System 3 is a conventional surface water treatment facility utilizing rapid mix, dual flocculation, sedimentation (tube settlers), chlorine oxidation, dual media filtration (anthracite and sand) and chlorine disinfection. Raw water is obtained from a reservoir approximately 28 feet deep, with sampling depth ranging from 3 to 9 feet below water surface. Facility chemical feed includes poly aluminum hydroxyl sulfate (coagulation), sodium hydroxide (pH adjustment), chlorine gas (oxidation and disinfection) and sodium hexametaphosphate (corrosion control). Chlorine is added between sedimentation and filtration.

3.1.4 Water System 4

Water System 4 is a surface water treatment facility utilizing oxidation, upflow contact clarification, granular activated carbon (GAC) filtration, UV treatment and chlorine disinfection. Raw water is obtained from a combination of reservoir surface water and groundwater. Facility chemical feed includes potassium permanganate (oxidation of manganese), polyaluminum chloride (coagulation), cationic polymer (coagulant aid), sodium hypochlorite (oxidation and disinfection), lime (pH adjustment) and phosphate (corrosion control).

3.2 Test Results

Samples were collected by Comprehensive Environmental Inc. staff and delivered to the University of New Hampshire (UNH) Center for Freshwater Biology for analysis of microcystin using the EnviroLogix Quantiplate-ELISA Kit with increased sensitivity. The samples are concentrated 10X and calibrators are extended so the method detection limit in the water sample is 2.5 ng/L, well below the 160 ng/L LOD of the standard ELISA kit. Water samples for this study were first frozen and then lyopholized to dryness. The dried material was rehydrated with double-distilled water then filtered through a 0.2 micron Whatman filter. This process prepares the sample for ELISA analysis for total free microcystins.

Tables 3-1 through 3-4 and Figures 3-1 through 3-4 provide the results of testing for microcystins at various sampling points throughout the four water treatment facilities evaluated[3]. All of the samples are below the WHO recommended guideline for drinking water of 1.0 μg/L or 1,000 ng/L microcystins including the raw water samples. In most of the samples there is a measureable decrease in microcystins from the raw water, post select treatment processes and the finished water.

Table 3-1
Microcystin: Water System 1

	Microcystin (ng/L)						
Date	Combined Raw	River	Brook	Well	Clarified	Filtered	Finished
7/14/2010	13.1	48.19	18.6	2.6	10.18	3.51	BDL
7/28/2010	20.09	NS	NS	NS	10.3	BDL	BDL
8/4/2010	22.72	NS	NS	NS	14.35	8.29	7.76
8/11/2010	39.97	NS	NS	NS	27.36	9.81	10.86
8/18/2010	25.84	NS	NS	NS	18.98	8.36	BDL

NS = No Sample
BDL = Below Detection Limit of 2.5 ng/L

Table 3-2
Microcystin: Water System 2

	Microcystin (ng/L)		
Date	Raw	Filtered	Finished
7/23/2010	9.49	6.34	3.99
8/10/2010	18.68	9.03	BDL
9/24/2010	15.07	9.1	BDL

BDL = Below Detection Limit of 2.5 ng/L

[3] Note: UNH has found an average of approximately 15 ng/L in extensive samples from over 50 New Hampshire recreational and water supply lakes. Much higher levels are typically found in blooms or mats that may collect in some areas (personal communication, Dr. James Haney, UNH Center for Freshwater Biology).

Table 3-3
Microcystin: Water System 3

	Microcystin (ng/L)		
Date	Raw	Clarified	Finished
8/11/2010	9.17	BDL	BDL
8/18/2010	5.7	BDL	BDL
8/25/2010	9.93	BDL	BDL
9/1/2010	15.69	4.22	BDL
9/8/2010	16.23	BDL	BDL

BDL = Below Detection Limit of 2.5 ng/L

Table 3-4
Microcystin: Water System 4

	Microcystin (ng/L)				
Date	Combined Raw	Wells	Pond	Filtered	Finished
9/16/2010	10.92	BDL	13.26	10.39	BDL

BDL = Below Detection Limit of 2.5 ng/L

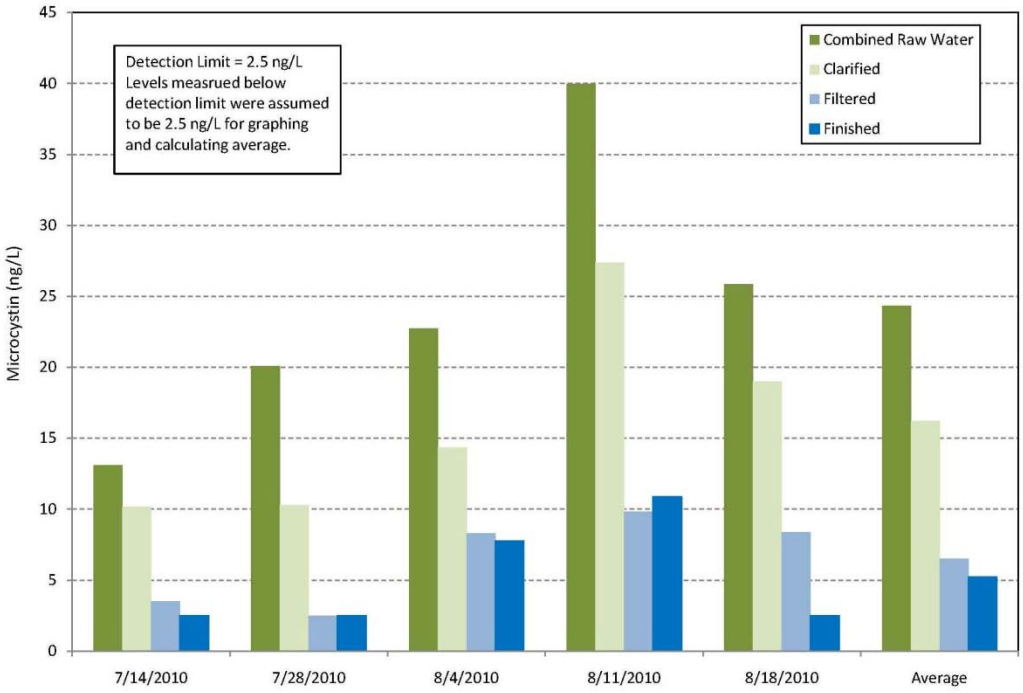

Figure 3-1
Microcystin: Water System 1

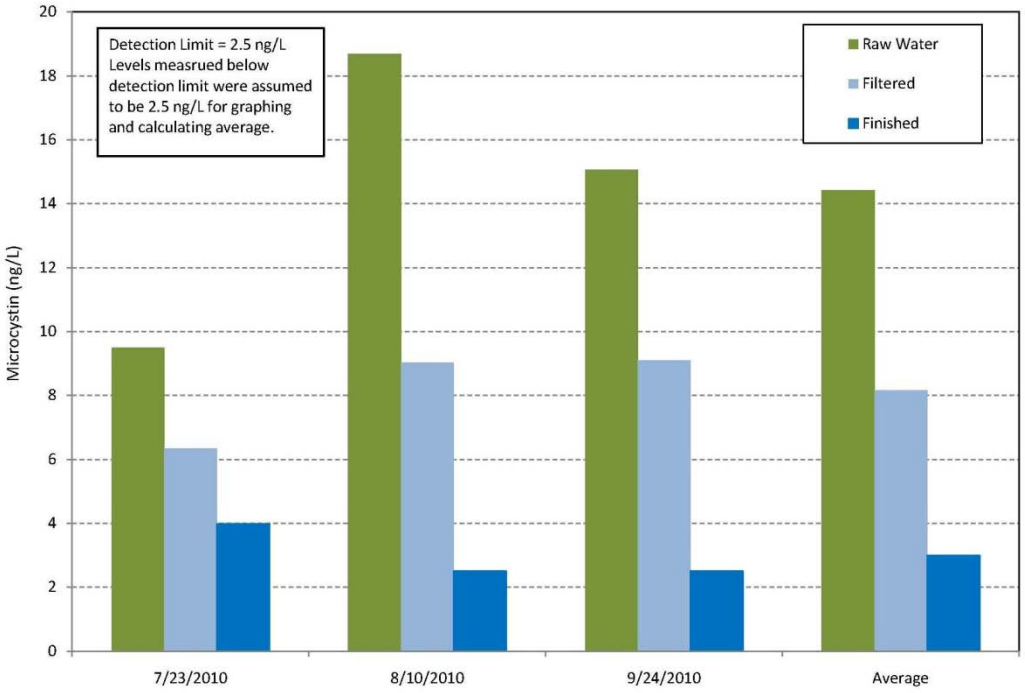

Figure 3-2
Microcystin: Water System 2

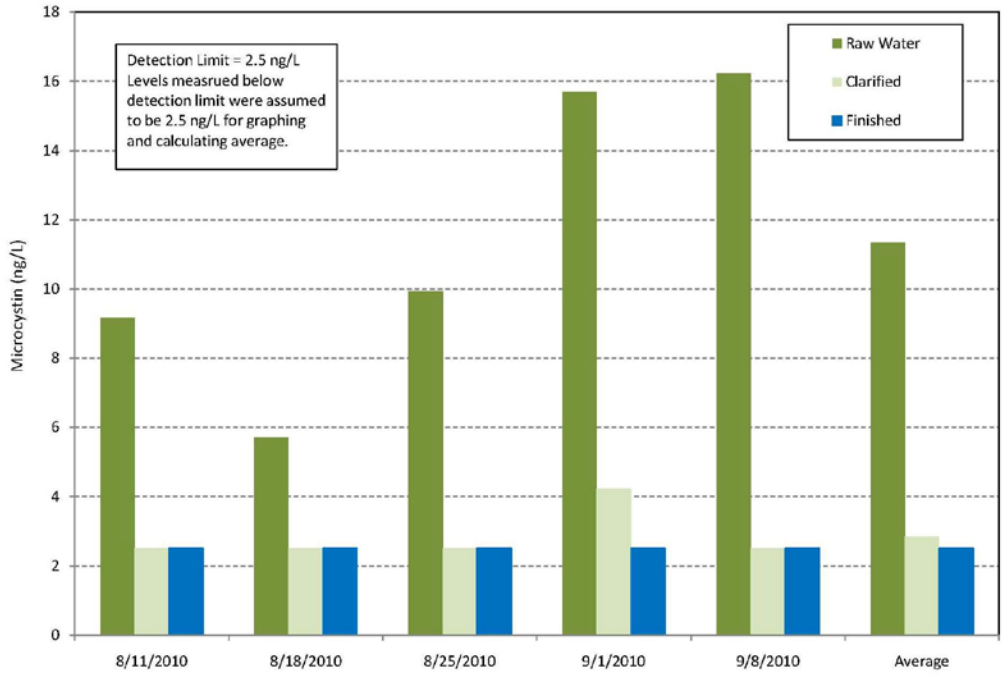

Figure 3-3
Microcystin: Water System 3

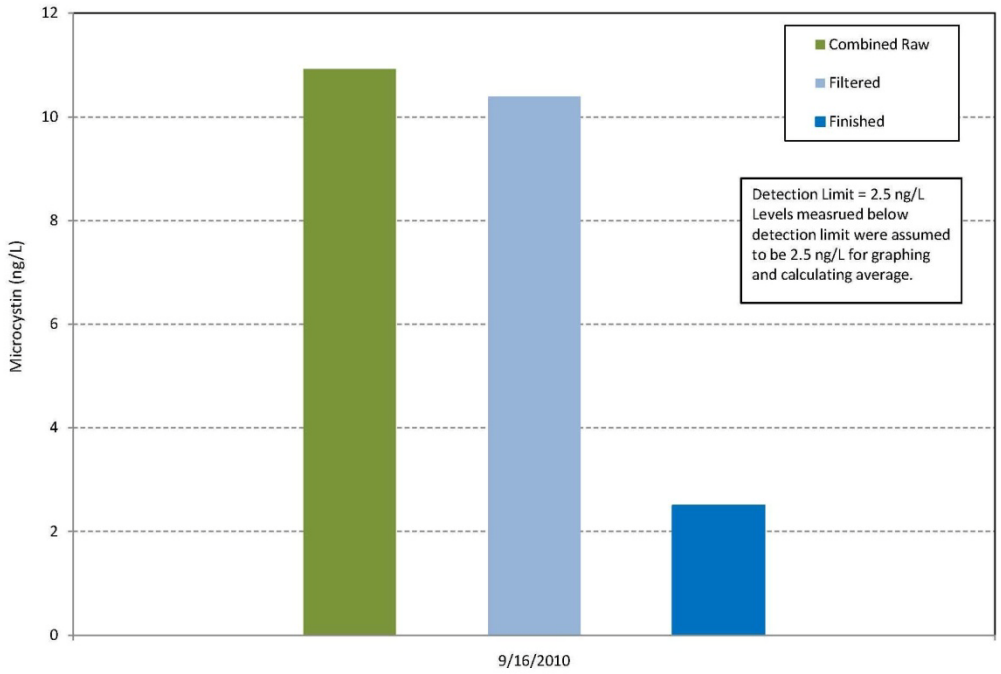

Figure 3-4
Microcystin: Water System 4

3.3 Data Analysis

While limited data were collected, the data seem to confirm some of the literature regarding the ability of various treatment processes to removal microcystins. Table 3-5 presents the microcystin removal percentages provided by each water treatment facility. Figure 3-5 provides a graphical representation of the average microcystin removal percentages for each water treatment facility. Conventional treatment (clarification/filtration) was successful in reducing a high percentage of microcystins, presumably intracellular microcystins. Microcystin removal percentages ranged from 33% to >88% for conventional treatment (clarification followed by filtration). Each of the evaluated systems disinfect using chlorine, with finished water microcystin removal percentages ranging from >56% to >90%. The removal percentages are calculated from the ratio of the microcystin concentration in the raw water and treated water. Note that microcystin levels were all low in the raw waters during the study and no bloom conditions were identified. What is not known is how the treatment processes would respond to blooms that enter the process as a slug, but some operators have noted a black scum on filters that may be indicative of cyanobacteria.

Table 3-5
Microcystin Removal Percentages from Raw Water

	Water System 1			Water System 2		Water System 3		Water System 4	
Sample	Clarified	Filtered	Finished	Filtered	Finished	Clarified	Finished	Filtered	Finished
1	22%	73%	>81%	33%	58%	>73%	>73%	5%	>77%
2	49%	>88%	>88%	52%	>87%	>56%	>56%	--	--
3	37%	64%	66%	40%	>83%	>75%	>75%	--	--
4	32%	75%	73%	--	--	73%	>84%	--	--
5	27%	68%	>90%	--	--	>85%	>85%	--	--
Average	33%	73%	79%	41%	76%	>72%	>74%	5%	>77%

Samples measured BDL assumed to be the detection limit of 2.5 ng/L in order to calculate removal percentages actual removals are greater than the value shown.
Water System 1: Super Pulsator Clarification, Filtration, Chlorine Disinfection
Water System 2: Sedimentation, Filtration, Chlorine Disinfection
Water System 3: Tube Settlers, Chlorination, Filtration
Water System 4: Oxidation, Upflow Contact Clarification, GAC, UV treatment, Chlorine Disinfection

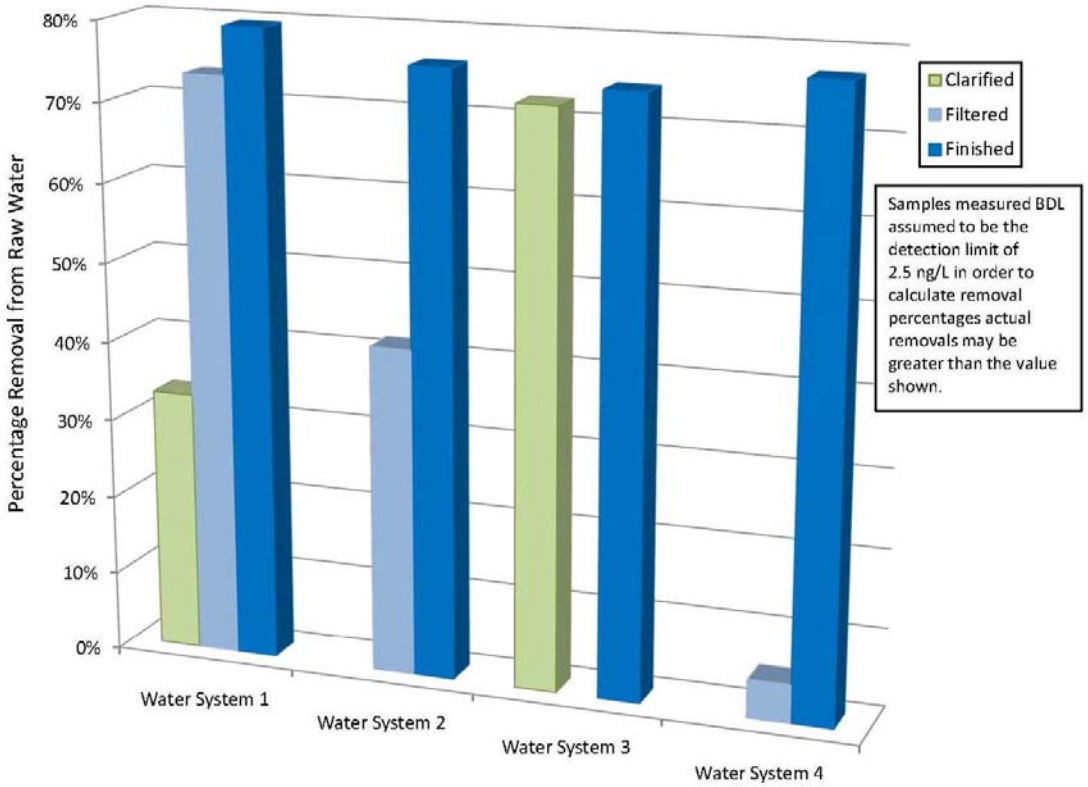

Figure 3-5
Average Microcystin Removal Percentages from Raw Water

The levels of microcystin in the finished water and raw water of each facility were well below the WHO recommended guideline for drinking water of 1,000 ng/L (1 μg/L) during this study. Additional data are needed to make a determination as to the ability of each facility's processes to successfully remove microcystins. Higher concentrations of microcystins in the raw water would help to evaluate the robustness of each process and facility.

Section 4.0
Recommended Actions

4.1 Monitoring and Testing

Typically, the first detection of a cyanobacteria bloom occurs visually. The observer may be a watershed inspector or a patron of a recreational waterbody. Surface water suppliers may consider training staff and volunteers to recognize and report cyanobacteria blooms. For recreational waters, public education may strengthen the community's role as first responders.

For waterbodies that commonly harbor algae and cyanobacteria blooms, regular sampling and testing helps to characterize seasonal and annual trends. A sampling program may be streamlined by including locations where blooms frequently occur as priority sample sites. Testing for cyanobacteria may be completed in the field or laboratory. Testing for cyanotoxins may be done onsite, or could be completed in the laboratory and depending on the level of accuracy needed.

Several other constituents of surface water may indicate the likelihood of a cyanobacteria bloom indirectly including dissolved oxygen, pH, total phosphorus, chlorophyll *a*, phycocyanin, counts of algae and phytoplankton. If a bloom does occur, monitoring these parameters as well as cyanotoxins throughout the lifespan of the bloom will help to characterize patterns typical of the waterbody. Fluctuations in these parameters and cyanobacteria blooms often occur with changes in weather or season. Comprehensive monitoring, therefore, allows some predictability for future prevention or mitigation efforts.

The development of a hydrologic and nutrient budget for each water body, including bathymetric mapping, is also helpful in eliminating potential sources of phosphorus and managing the watershed. Intake relocation to avoid areas where cyanobacteria usually accumulate may also be a possible option for some water bodies.

4.1.1 Tests for Cyanobacteria and Cyanotoxins

Water may be analyzed on site for cyanobacteria with field instruments such as the portable sensor for phycocyanin fluorescence available from Yellow Springs Instruments (YSI). The instrument detects cells by the fluorescence of a pigment unique to cyanobacteria, phycocyanin. The instrument is also capable of detecting chlorophyll *a*, present in both cyanobacteria and algal groups, which may help to differentiate algae from cyanobacteria. For comprehensive monitoring, this instrument can be used from a boat to sample along a route or in specific locations. Following transects and sampling at various depths will show whether a surface water intake can be moved to avoid cyanobacteria blooms. Real-time camera monitoring, such as the FlowCAM, provides another option for on-site monitoring. With this method, a camera monitors particles in fluid stream to count, image and analyze cells in discrete sample or continuous flow.

Methods for measuring some cyanotoxins include: enzyme-linked immunosorbent assays (ELISA), protein phosphatase inhibition assays (PPIA), high performance liquid chromatography (HPLC) and liquid chromatography – mass spectrometry (LC/MS). The method selected will depend on the type of cyanotoxin to be monitored, new developments in laboratory methodologies and federal and state approval of select methodologies for use in drinking water analysis.

Several kits are available for analyzing samples for microcystin in the field and laboratory, but most require a step to release cyanotoxins by breaking the cells (lysis) via freeze-thaw cycles which may be difficult without a full laboratory. EnviroLogix, Abraxis, Enviroguard and Beacon kits are available for microcystin testing both within the field and laboratory. ELISA test kits for cylindrospermopsin are also available, but there are no ELISA test kits for anatoxin-a.

Water samples can be tested for cyanobacteria and cyanotoxins in the field or by laboratories. However, grab sampling methods are subject to the presence of cyanobacteria and/or cyanotoxins in the water at the time of sampling. There is a possibility of missing water containing cyanobacteria and/or cyanotoxins using the grab sample technique. In-line monitoring equipment is available for continuous

monitoring of cyanobacteria. Phycocyanin blue-green algae sensors can be installed to monitor for cyanobacteria. These sensors measure the fluorescence of phycocyanin in the living cyanobacteria cells. In-line monitoring can provide operators with an early warning system for increasing cyanobacteria biomass that may indicate a potential taste and odor event and/or cyanotoxin increase.

4.1.2 Developing a Monitoring Plan

Both visual monitoring and sample testing can contribute to a comprehensive monitoring plan. Steps in a plan logically proceed from the immediate, first response to the in-depth analysis: visual inspection, water quality testing, cyanobacteria sampling, and finally cyanotoxin analysis. The following provides recommended steps in monitoring cyanobacteria levels, adapted from *Toxic cyanobacteria in water: A guide to their public health consequences, monitoring and management,* World Health Organization, 1999.

Step 1 – Visual Site Inspection (Weekly or Two-Weekly Intervals)

- If discoloration, scums or mats indicate cyanobacteria perform Step 3 and Step 2.
- If transparency is less than 2 meters, discolored or turbid perform Step 2.

Step 2 – Monitor Total Phosphorus

- If total phosphorus concentrations are below 0.01 to 0.02 mg/L mass developments of cyanobacteria are unlikely and high turbidities may have other causes.
- If concentrations of total phosphorus are higher:
    - Perform Step 3
    - Consider monitoring further nutrients and hydrological parameters
    - Inspect the catchment for the source of the problem.

Step 3 – Monitor Mass Developments of Cyanobacteria (Two-Weekly Interval)

- If levels exceed selected alert values:
    - Take action to protect health
    - Intensify monitoring to weekly or more frequently
    - Perform Step 4
    - Consider collecting supplementary information

Step 4 – Monitor Toxin Content of Cyanobacteria

- If toxin levels exceed guideline values and conditions, confirm the need for action:
    - Monitoring finished water cyanotoxins
    - Implementing additional treatment measures

Table 4-1, from the abovementioned WHO report, shows potential parameters and related logistics for planning various monitoring efforts. Note that monitoring and identifying cyanobacteria can be completed by local personnel managing the waterbody with specific training. Quality control can then be done periodically by a nearby lab or expert.

In formulating a monitoring plan, consider whether the history of the waterbody prompts a more proactive protocol. The following should be considered in selecting sample sites:

- Is the waterbody used primarily for recreation? Focus sampling at shorelines.
- Is the waterbody used primarily for drinking water? Prioritize sampling near intake.
- Determination of total cyanobacteria population requires a central reference (open, mixed water) with samples taken at various depths to characterize strata.

Table 4-1
Approaches to Monitoring for Cyanobacteria and Analysis for Cyanotoxins
(Source: *Toxic cyanobacteria in water: A guide to their public health consequences, monitoring and management*, World Health Organization, 1999)

Monitoring type	Parameters/variables	Demands on equipment and skills	Who	Where
Basic		Minimal		
Site inspection for indicators of toxic cyanobacteria in waterbody	Transparency, discolouration, scum formation, detached mat accumulation	Secchi disc, regular site inspection by trained staff; skill requirement basic, training easily provided	Environmental or health officers, trained health staff or supervised local	Local
Background		Low to moderate		
Potential for cyanotoxin problems in waterbody	Total phosphorus, nitrate and ammonia, flow regime, thermal stratification, transparency	Photometer, boat, depth sampler, Secchi disc, submersible temperature/oxygen probe; skills basic but require specific training and supervision	Environmental officers or experts with limnological expertise	Local, regional
Cyanobacteria		Low to moderate		
In waterbody and drinking water	Dominant taxa (quantity): often determination to genus level only is sufficiently precise; quantification only as precise as needed for management	Microscope, photometer is useful; specific training and supervision is required, but quite easily achieved	Environmental or health officers (with occasional quality control by experts); consultants with limnological expertise	Local, regional
Toxicity assessment		Moderate		
In waterbody and drinking water	Toxicity	Demands on equipment are low, but rather high on skills	Toxicologists	Central
Toxin concentration		Moderate to high		
In waterbody and drinking water	Toxin concentration	New methods with lower financial demands presently in development for some cyanotoxins (e.g. immuno-assay); skill requirements vary widely from moderate to very high	Skilled analysts	Central

4.2 Preventative Actions

Preventative actions are the first barrier in the multiple barrier approach to supplying safe drinking water. By preventing cyanobacteria growth within the drinking water supply, operators will not need to rely on treatment removal methods. Watershed management is the most effective measure for limiting the growth of cyanobacteria. Water resource protection and management methods, as presented in Section 2.0 of this report, involve limitation of nutrient loading from surface runoff and erosion, stormwater discharge and wastewater discharge.

4.3 Mitigation

Cyanobacteria can be removed through methods taken at the source or treatment facility. Removal and inactivation of cyanobacteria and intracellular and extracellular cyanotoxins requires a combination of processes or multiple barrier approach as described in Section 2.0 of this report. Source cyanobacteria control can be accomplished through nutrient precipitation, dredging, use of algaecides, and use of ultrasonic sound wave equipment. Each treatment facility is unique and may need to be examined specifically for cyanobacteria and cyanotoxins. Intracellular cyanotoxins may be removed by conventional treatment and membrane filtration. Extracellular cyanotoxins may be degraded by chlorine at varying pH values.

4.4 Further Investigations

The levels of microcystin in the finished water and raw water of each facility included in this study were well below the WHO recommended guideline for drinking water of 1,000 ng/L. Additional data is needed to make determination as to the ability of each facility's processes to successfully remove microcystins. Higher concentrations of microcystins in the raw water would help to evaluate the robustness of each process and facility. Further investigations are needed to monitor the concentrations of microcystins in raw water supplies and finished water.

Research of cyanobacteria and cyanotoxins is ongoing. There is a fair amount of data available regarding microcystins and the ability of various treatment processes to remove microcystins. There are a number of other cyanotoxins about which much less is known.

It will be necessary to be aware of ongoing research and communicate those results to the drinking water community.

The USEPA is evaluating cyanotoxins, their potential health risks, the ability to reasonably treat them and whether there is a need to regularly test for them. The USEPA anticipates making a determination regarding three cyanotoxins by 2013.

Further research and investigation into cyanotoxins is needed, with the aim to answer the following questions:

- What are the true health risks associated with ingestion of various cyanotoxins in drinking water at low levels over the long-term?
- Since cyanotoxin levels can fluctuate greatly over brief periods of time, what are the actual concentrations of cyanotoxins in raw water supply sources over days, weeks and months?
- What is the true ability of treatment processes to remove various cyanotoxins including microcystins?
- Are groundwater sources susceptible to cyanobacteria and if so at what levels?
- What steps should be taken if the system if flooded with bloom slugs or mats that coat processes and may affect operations?
- What would a comprehensive management program for cyanobacteria look like?

Appendix A
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