# TENMILE LAKES TOXIC ALGAL SAMPLING PROGRAM: 2007 DATA SUMMARY REPORT


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## BACKGROUND

As a continuation of previous toxic algal monitoring, Tenmile Lakes were sampled in 2007 to assess the dynamics of the potentially toxigenic blue-green algal species, *Microcystis aeruginosa* and various *Anabaena* species. *Microcystis* produces hepatotoxins (known as microcystins), and *Anabaena* produces both neurotoxins (anatoxin-a) and microcystin. Both toxins are capable of harmful effects to animals and humans (Chorus and Bartram 1999). A toxic bloom of *M. aeruginosa* was first documented in Tenmmile Lakes in September of 1997, prompting the Oregon Department of Health to issue a health advisory recommending that the lakes not be used for drinking water and that contact recreation be avoided (Kann and Gilroy 1997). The goal of 2007 sampling, performed by the Tenmile Lakes Basin Partnership, was to determine presence and cell density of these potentially toxic species at a limited number of sampling stations. Cell density of potentially toxigenic species was then compared to drinking water guidance levels for lakes and reservoirs (e.g., Yoo et al. 1995; Chorus and Bartram 1999).

## METHODS

Four stations (2 in each lake) were sampled to cover a major arm and open-water location in each lake (Fig. 1). Stations S8 and N16 are centrally located and S3 and N11 are located near the terminus of Templeton Arm and Big Creek Arm, respectively. These stations were sampled 8 times beginning June 25th and ending September 25th, 2007.

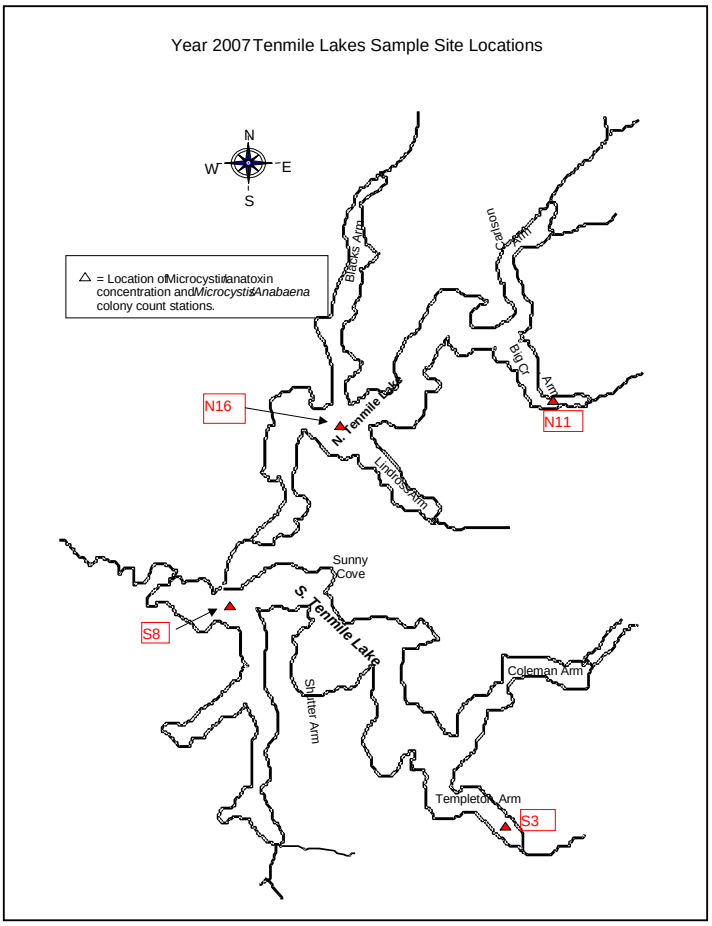


**Figure 1. Location of standard toxic algal sampling stations in Tenmile Lakes, 2007.**

Because the goal of the *M. aeruginosa* and *A. flos-aquae* sampling was to detect conditions that may pose human health hazards, samples were collected mid-day and integrated over the upper 1/3 of the water column at the open-water stations (S8 and N16), and over the entire water column at the shallow stations (S3 sand N11). At each of the established sampling locations a vertical tow ranging between 1 to 2.5 meters of the water column (depending on location) was made using a 64-µm plankton net.

The filtered contents of 3 replicate hauls were composited in a bucket and then placed in a 250 ml opaque sample bottle containing 1% Lugol's preservative and shipped to plankton taxonomist Jim Sweet of Aquatic Analysts, INC., who performed a microscopic analysis for *Microcystis* and *Anabaena* density (cells $ml^{-1}$). For toxin analyses the contents of 3 replicate hauls taken with a tube sampler were composited in a bucket and then placed in a 1 liter bottle with no preservative and frozen at the TLBP office. If counts received the following week from Aquatic Analysts, INC showed that cell counts considerably exceeded the Alert Level 2 threshold of 2000 cells $ml^{-1}$, the frozen samples were then shipped overnight air on ice to CyanoLab (division of GreenWater Labs in Palatka, FL). The enzyme linked immunosorbent assay (ELISA) was then used to determine microcystin toxin and LC/MS to determine anatoxin-a (Note: because health advisories and media outreach are initiated based upon cell density and not toxin concentration, toxin analysis is not prioritized when budgetary constraints exist or when cell counts are below 15,000 cells/ml).

Also in 2007, two sets of samples of treated tap water and raw lake intake water were collected on 8/7 and 10/27 with help from an anonymous Tenmile Lake homeowner. In addition, a duplicate sampling tow for quality assurance was collected on two occasions (station D on attached lab sheets), and an additional sample on 7/9 was re-sampled to assess duplicate variability.

## RESULTS

### *2007 Trends*

Samples from the first sample trip of June 25th show that cell density of potentially toxigenic cyanobacteria species in South Tenmile (stations S3 and S8) did not exceed the WHO Alert Level 1 guideline of 500 cells ml-1 (Yoo et al. 1995; also known as the increased vigilance level, Table 1; Figure 2) for either *Microcystis aeruginosa* (MSAE) or total *Anabaena*. However, in north Tenmile Lake, cell density of total *Anabaena* exceeded Alert Level 1 at both N11 and N16, and the combined count of MSAE and Gloeotrichia (GTEC) just exceeded (2085 cells/ml) the Alert Level 2 guideline of 2000 cells/ml at N16 (Alert Level 2 is when DHS and local health services typically issue a public alert for drinking water lakes and reservoirs). The combined density of MSAE and GTEC is computed because GTEC is also potential microcystin producer (Carey et al. 2007).

On July 9th *MSAE* exceeded the World Health Organization (WHO) Alert Level 2 guideline of 2000 cells $ml^{-1}$ only at station S8 (2529 cells/ml; Figure 2; Table 1); while total *Anabaena* increased at all stations, but remained below 2000 cells/ml. On the subsequent three sample dates (7/23, 8/6, and 8/22) MSAE cell density declined and remained at less than 1000 cells/ml; however, total *Anabaena* continued to increase at most stations, with a maximum of 5207 cells/ml at station N16 (Figure 2; Table 1). MSAE density increased at both of the stations located in the arms (S3 and N11; Figure 1) on September 12th, with S3 exceeding the WHO Alert Level 1 guideline, and N11 at 6,605 cells/ml exceeding the WHO Alert Level 2 guideline of 2,000 cells/ml. Levels of total *Anabaena* remained similar to the previous sample period at both N11 and N16; with N16 continuing to exceed the Alert

**Table 1. Tenmile Lakes Algal Count and Toxin Results, 2007 (nt=not tested; toxin lab results in Appendix II)**

| *Station* | *Date* | *Microcystis aeruginosa (cells/ml)* | *Gloeotrichia echinulata (cells/ml)* | *Microcystis + Gloeotrichia (cells/ml)* | *Anabaena flos-aquae (cells/ml)* | *Anabaena planktonica (cells/ml)* | *Anabaena circinalis (cells/ml)* | *Anabaena sp. (cells/ml)* | *Total Anabaena (cells/ml)* | *Microcystin (ug/L)* | *Anatoxin (ug/L)* |
|---|---|---|---|---|---|---|---|---|---|---|---|
| S3 | 6/25/2007 | 47 | 0 | 47 | 321 | 0 | 0 | 0 | 321 | nt | nt |
| S8 | 6/25/2007 | 349 | 0 | 349 | 24 | 67 | 0 | 0 | 91 | nt | nt |
| N11 | 6/25/2007 | 0 | 706 | 706 | 281 | 353 | 0 | 0 | 634 | nt | nt |
| N16 | 6/25/2007 | 1267 | 818 | 2085 | 310 | 297 | 0 | 0 | 607 | nt | nt |
| S3 | 7/9/2007 | 702 | 0 | 702 | 202 | 296 | 0 | 0 | 498 | nt | nt |
| S8 | 7/9/2007 | 2529 | 0 | 2529 | 357 | 1024 | 0 | 0 | 1381 | nt | nt |
| N11 | 7/9/2007 | 127 | 993 | 1120 | 669 | 791 | 0 | 0 | 1460 | nt | nt |
| N16 | 7/9/2007 | 0 | 0 | 0 | 883 | 474 | 0 | 0 | 1357 | nt | nt |
| S3 | 7/23/2007 | 0 | 0 | 0 | 35 | 818 | 0 | 0 | 853 | nt | nt |
| S8 | 7/23/2007 | 12 | 0 | 12 | 205 | 1746 | 0 | 0 | 1951 | nt | nt |
| N11 | 7/23/2007 | 159 | 1124 | 1283 | 754 | 1184 | 0 | 0 | 1938 | nt | nt |
| N16 | 7/23/2007 | 831 | 0 | 831 | 489 | 4718 | 0 | 0 | 5207 | nt | nt |
| S3 | 8/6/2007 | 107 | 0 | 107 | 71 | 3293 | 0 | 0 | 3364 | nt | nt |
| S8 | 8/6/2007 | 369 | 0 | 369 | 0 | 4795 | 0 | 0 | 4795 | nt | nt |
| N11 | 8/6/2007 | 450 | 0 | 450 | 0 | 109.7 | 0 | 0 | 110 | nt | nt |
| N16 | 8/6/2007 | 866 | 0 | 866 | 23 | 3620 | 0 | 0 | 3643 | nt | nt |
| S3 | 8/22/2007 | 223 | 0 | 223 | 0 | 1179 | 0 | 0 | 1179 | nt | nt |
| S8 | 8/22/2007 | 113 | 0 | 113 | 0 | 4995 | 0 | 0 | 4995 | nt | nt |
| N11 | 8/22/2007 | 318 | 0 | 318 | 12.3 | 286 | 0 | 0 | 298 | nt | nt |
| N16 | 8/22/2007 | 464 | 0 | 464 | 0 | 2851 | 14 | 0 | 2865 | nt | nt |
| S3 | 9/12/2007 | 1877 | 0 | 1877 | 0 | 3162 | 0 | 0 | 3162 | nt | nt |
| S8 | 9/12/2007 | 131 | 0 | 131 | 0 | 8687 | 0 | 0 | 8687 | nt | nt |
| N11 | 9/12/2007 | 6605 | 0 | 6605 | 0 | 304 | 0 | 0 | 304 | 5.4 | nt |
| N16 | 9/12/2007 | 69 | 0 | 69 | 0 | 2818 | 0 | 0 | 2818 | nt | nt |
| S3 | 9/25/2007 | 1632 | 0 | 1632 | 0 | 227 | 0 | 0 | 227 | nt | nt |
| S8 | 9/25/2007 | 1115 | 0 | 1115 | 0 | 1274 | 0 | 0 | 1274 | nt | nt |
| N11 | 9/25/2007 | 2771 | 0 | 2771 | 0 | 227 | 0 | 0 | 227 | 7.5 | nt |
| N16 | 9/25/2007 | 277 | 0 | 277 | 68.6 | 4221.3 | 0 | 0 | 4289.9 | nt | nt |

Level 2 guideline (Figure 2). However, increases in total *Anabaena* occurred at both south-Lake stations, with S8 sharply increasing to 8,687 cells per ml (exceeding the Alert Level 2 guideline by 4.3 times). However, as shown in Table 1, the predominant species was *Anabaena planctonica*, a species less commonly associated with toxin production. On the final sample date of 9/25, Alert Level 2 continued to be exceeded at N11 for MSAE (2771 cells/ml), and at N16 for total *Anabaena* (Figure 2).

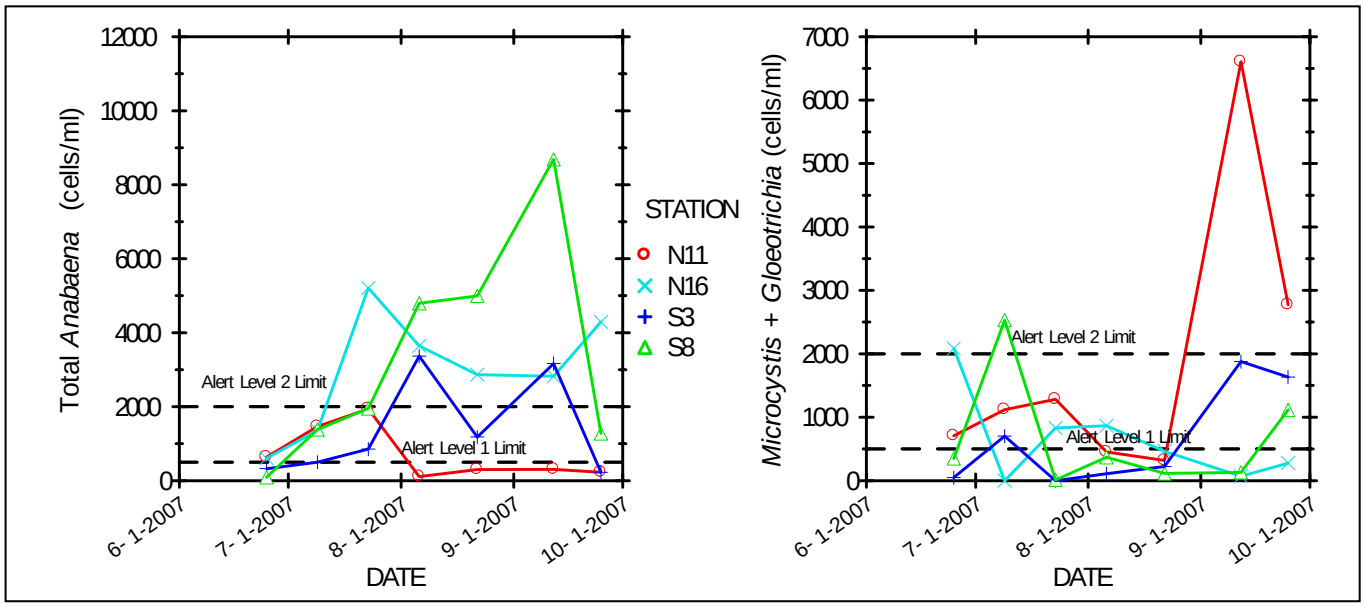


**Figure 2. Cell Density of *Microcystis aeruginosa* and total *Anabaena sp.* in Tenmile Lakes, 2007.**

### *2007 Algal Toxin Samples*

ELISA testing performed on the Sep 12th N11 sample containing 6605 cells/ml MSAE showed a microcystin concentration of 5.4 µg/L (Table 1). This value exceeded the drinking water microcystin standard of 1 µg/L by 5.4x. Subsequent microcystin analysis on 9/25 showed a slightly higher concentration of 7.5 µg/L even though MSAE density declined to 2771 cells/ml. However, as noted below, resolution of cell density measurements when species are non-dominant may be as much as ± several thousand cells/ml, and such discrepancies may also be due to the fact that the toxin samples are collected from raw water while the cell density samples are concentrated with a plankton net. Nonetheless, because past research from Tenmile Lakes as well as other lake systems indicates that 5000 cells/ml of MSAE is generally associated with 1 µg/L of the hepatotoxin microcystin (Jacoby and Kann 2007), it is unclear why microcystin was substantially higher than expected based on cell density (especially when other potential microcystin producers such as *Anabaena* were also minimal at this station; 304 and 227 cells/ml on 9/12 and 9/25). It is possible that much of the toxin was extra-cellular and due to release associated with higher counts nearby (e.g., blue-green blooms show a high degree of patchiness). Toxin samples measured in the lake on 10/27 showed a more typical relationship of ~1 µg/L microcystin for ~5000 MSAE cells/ml (see Table 3 below). Regardless of the cause, 7.5 µg/L of microcystin greatly exceeds the 1 µg/L drinking water standard and was close

to the recreational standard of 8 ug/L (Stone and Bress 2007). For homeowners who are drawing water to treat for potable uses it is essential to reduce toxin levels down to or below 1 ug/l.

These data show that although cell density levels were still relatively low with respect to recreational guidelines (e.g., which would be 100,000 cells/ml for *Anabaena* and 40,000 cells/ml for MSAE; Stone and Bress 2007) that the potential to exceed drinking water standards and to approach recreational guidelines exists even when MSAE cell density is the range of 2500-6500 cells/ml.

### *Sampling Resolution Issues*

As noted in the methods, a duplicate plankton tow was collected on two occasions in 2007 to assess tow reproducibility and potential non-uniform cell density distribution at a station. These results show that on 6/25 the difference between the original sample and the duplicate was low for total *Anabaena* and was 1134 cells/ml for MSAE, but that both samples were similar with respect to public health alert levels (Table 2).

**Table 2. Difference in cell density between original sample tow and duplicate sample tow for MSAE and *Anabaena* in Tenmile Lakes, 2007.**

| *Station* | *Date* | *Aphanizomenon flos-aquae1* | *Microcystis aeruginosa (cells/ml)* | *Anabaena flos-aquae (cells/ml)* | *Anabaena planktonica (cells/ml)* | *Anabaena circinalis (cells/ml)* | *Anabaena sp. (cells/ml)* | *Total Anabaena (cells/ml)* | *MSAE sample-duplicate difference* | *Total Anabaena sample-duplicate difference* |
|---|---|---|---|---|---|---|---|---|---|---|
| N16 | 25-Jun | 1,910 | 1,267 | 310 | 297 | 0 | 0 | 607 | | |
| N16-D | 25-Jun | 2,523 | 133 | 406 | 571 | 0 | 0 | 977 | 1,134 | -370 |
| S8 | 9-Jul | 10,203 | 2,529 | 357 | 1,024 | 0 | 0 | 1,381 | | |
| S8-D | 9-Jul | 13,509 | 0 | 63 | 991 | 0 | 0 | 1,054 | 2,529 | 327 |
| S8-Archive | 9-Jul | 10,652 | 649 | 210 | 431 | 0 | 0 | 1,290 | 1,880 | 91 |
| | 9-Jul Mean | 11,455 | 1,059 | | | | | 1,242 | | |
| | 9 Jul ±90% CI | 3,023 | 2,214 | | | | | 618 | | |

On 7/9 the sample-duplicate comparison showed 2529 cells/ml MSAE in the original sample; however, MSAE was not detected in the duplicate sample (Table 2). An archived split of the original sample was then analyzed, showing an MSAE level of 649 cells/ml. Such differences are likely due to large relative errors when a species is non-dominant (e.g., MSAE represented only 0.8% of the total algal density on 7/9) and the difference between samples can be due to observing ± only one or two colonies during microscopic analysis. Thus, for non dominant species, reported levels indicate the general trend but do not guarantee that low levels of potentially toxigenic species are not present in the lake. For more dominant species such as *Aphanizomenon* (73% of total density on 7/9) or total *Anabaena* (7.5% of total density on 7/9), sample-duplicate differences were well within acceptable limits (Table 2). These results underscore both the patchiness that algal densities exhibit in lake environments and limited resolution of laboratory determinations of algal density when species are not dominant.

### *Sampling of Home Water Treatment System*

Similar to data from 2006, cell density data from a home water treatment system showed that *Anabaena* cells were detected in samples collected directly from treated kitchen tap water (Table 3). On August 7th, 27,620 cells/ml of total *Anabaena* were found in the lake near the treatment system intake, with a reduction to 742 cells/ml in treated tap water (representing a 97.3% reduction). Although no *Anabaena* cells were detected at the lake intake on Oct 27th, 35 cells/ml were found in treated tap water (Table 3).

Despite being found near the treatment intake on both dates, MSAE was not detected in treated tap water (Table 3). However, for other dominant species such as the diatom *Cyclotella* or the green alga *Chlamydomonas* only 55-70 percent of cells were removed on Oct 27th (see lab data sheet for Lake B and Kitch A on 10/27; Electronic Appendix I). Although these species are not toxigenic, it underscores the potential for high tap water concentrations of toxigenic algae should large in-lake blooms of such species develop.

Microcystin results for 10/27 show a concentration of 0.7 µg/L associated with the 4271 MSAE cells/ml in the lake near the intake to the home, and a non-detect in treated tap water (Table 3). Thus, at least for a low microcystin concentration the water treatment system was effective at reducing microcystin. However, given in-lake toxin concentrations >5x the drinking water standard of 1 µg/L (see above), it is essential that additional testing be performed on treatment system efficacy and adjustments made to treatment protocol if warranted.

**Table 3. Comparison of lake and treated tap water at Lindross Arm private residence, August 8yh and October 27th, 2007.**

| Lab Station ID | Description | Date | *Microcystis aeruginosa (cells/ml)* | *Anabaena flos-aquae (cells/ml)* | *Anabaena planktonica (cells/ml)* | *Anabaena circinalis (cells/ml)* | *Anabaena sp. (cells/ml)* | *Total Anabaena (cells/ml)* | *Microcystin (µg/L)* |
|---|---|---|---|---|---|---|---|---|---|
| A | Trt Tap | 8/7/2007 | 0 | 0 | 742 | 0 | 0 | 742 | |
| B | Lake intake | 8/7/2007 | 323 | 3839 | 23517 | 264 | 0 | 27620 | |
| A | Trt Tap | 10/27/2007 | 0 | 35 | 0 | 0 | 0 | 35 | Non-detect |
| B | Lake intake | 10/27/2007 | 4271 | 0 | 0 | 0 | 0 | 0 | 0.7 |

### *2002-2007 Comparison*

With the exception of 2002, density of MSAE in Tenmile Lakes in 2007 was somewhat higher than all previous years (Figure 3); and low numbers of MSAE were more consistently detected in 2007 than other years (see elevated lower quartile of the 2007 box plots; Figure 4). The consistent detection of low MSAE values may be due to a switch to higher counting resolution in 2007 (the number of total algal units microscopically counted was increased from 100 algal units to 200 algal units in 2007). Total *Anabaena* density was noticeably higher in 2007 than for the previous 2002-2006 period, with 9 incidences when cell density exceeded Alert Level 2 (Figures 3 and 4). However, the predominant *Anabaena* species was *Anabaena planktonica* (Table 1), a species less commonly associated with toxin production than *Anabaena flos-aquae*.

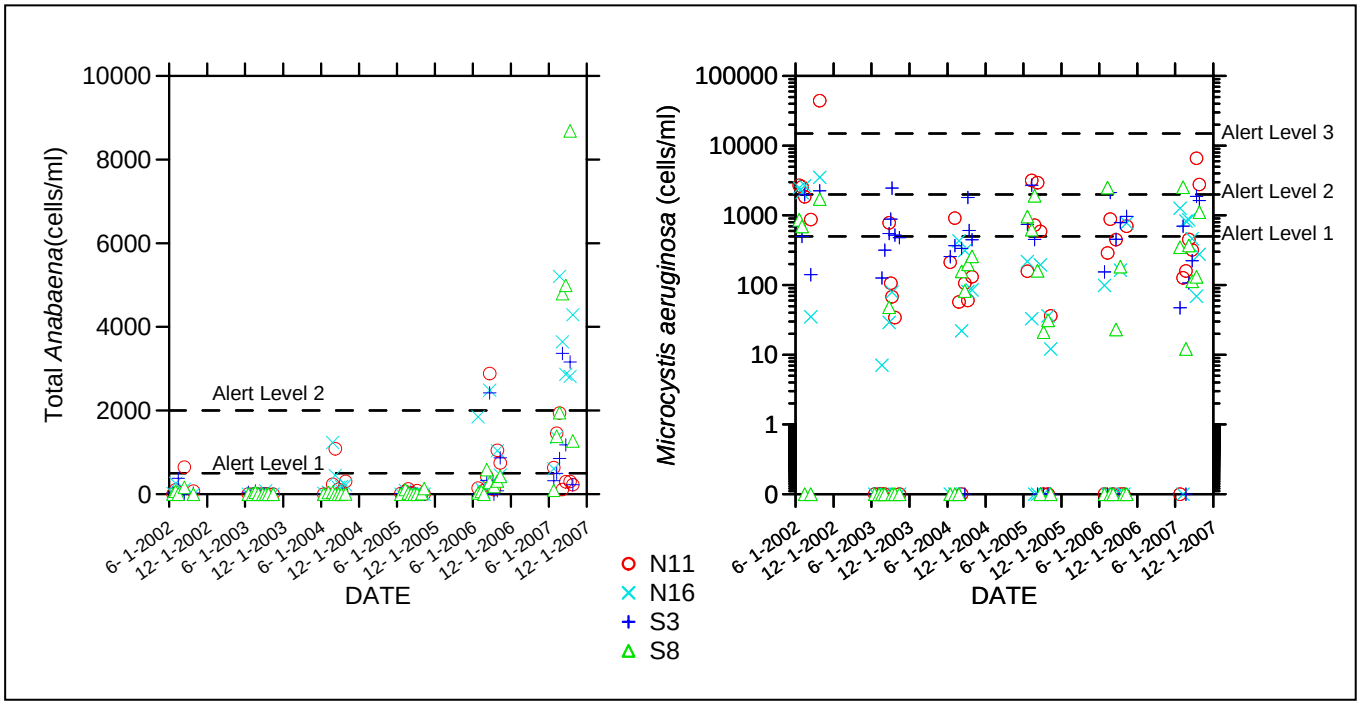


**Figure 3. Density of *Microcystis aeruginosa* and *Anabaena flos-aquae* in Tenmile Lakes, 2002-2007.**

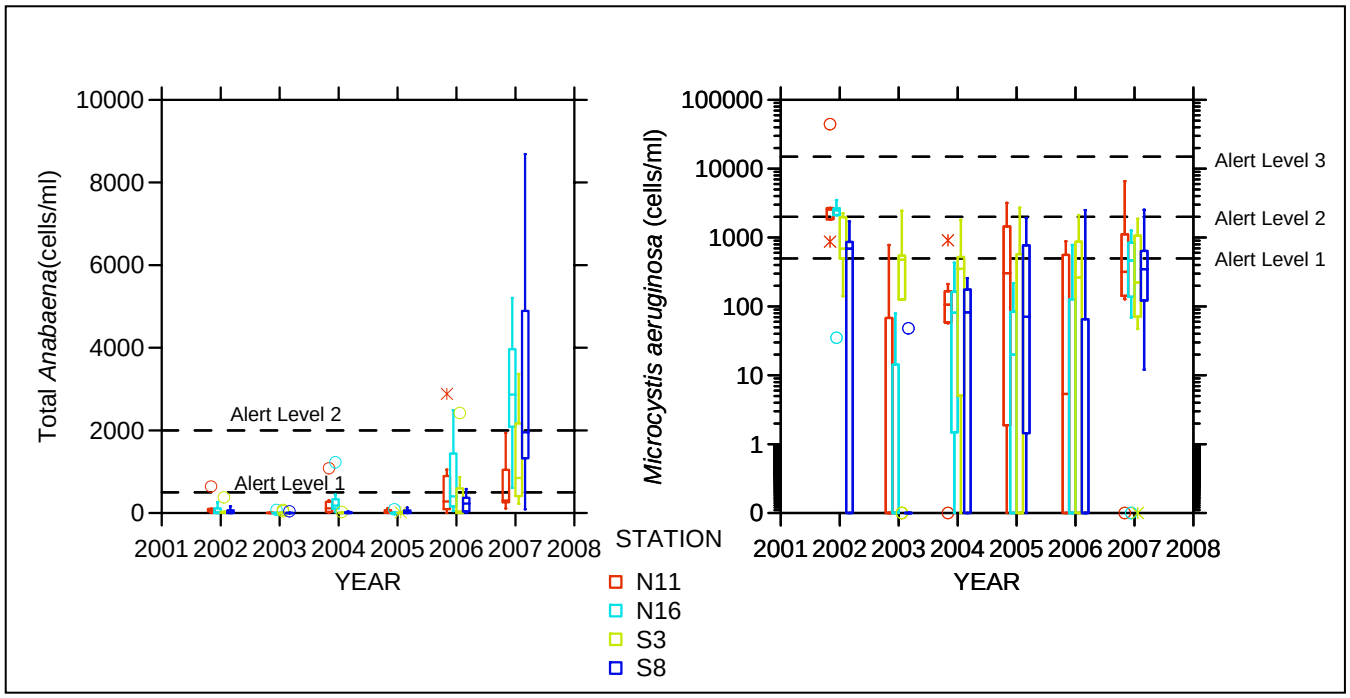


**Figure 4. *Microcystis aeruginosa* and *Anabaena flos-aquae* cell density grouped by station, Tenmile Lakes, 2002-2007.**

***Summary***

Although overall 2007 cell density values were low relative to recreational guidelines utilized by Oregon DHS (Stone and Bress 2007), there were numerous instances when either *Anabaena* or MSAE exceeded the Alert Level 2 drinking water guideline of 2000 cells/ml. Microcystin toxin analyses performed on lake samples in September showed values 5.4 to 7.5x the WHO drinking water guideline value of 1 µg/L. Moreover, the 7.5 µg/L microcystin result from 9/25 approached the 8 µg/L guideline value for recreational contact. These toxin levels occurred at MSAE cell density levels that were not substantially greater than the Alert Level 2 value of 2000 cells/ml. Thus, lake home-owners who utilize treated lake water for household purposes should always ensure their treatment systems are operational and up to date. This is again (similar results were shown in 2006) underscored by the above results showing low levels of *Anabaena* in treated tap water. Although microcystin testing on 10/27 showed a reduction from 0.7 µg/L to below detection in a home water treatment system , additional testing of treated tap water for algal toxins is essential for determination of treatment system efficacy at higher toxin concentrations and for homes with other treatment system configurations. Toxin results from 2007 reaffirm that drinking water concerns occur at much lower cell count and toxin levels than for generalized recreational contact advisories, and that toxin concentration may not always be associated with cell density at a particular location.

**Disclaimer**

Due to the patchy nature of blue-green algal blooms it is possible for higher *Microcystis* and *Anabaena* densities (and therefore higher microcystin or anatoxin concentrations) to be present in areas not sampled in this survey, particularly along shorelines or during calm conditions of little to no wind. Given the lakes' demonstrated history of toxic *Microcystis* and *Anabaena* blooms, and the fact that all areas of the lake cannot be tested at all times, those utilizing the lake for drinking water should always follow Oregon Health Division recommendations for purification. In addition, recreational users should always avoid contact with water whenever noticeable surface concentrations of algae are evident or when the lake has an obvious green to blue-green appearance. Moreover, because pets or other domestic animals are the most likely to ingest contaminated water, these animals should not be allowed access to the lakeshore whenever either noticeable surface concentrations of algae or an obvious green to blue-green appearance is evident.

A fact sheet about *Microcystis aeruginosa* and detailed recommendations for lake water treatment may be obtained from Ken Kauffman at 971-673-0435 or via E-mail at kenneth.w.kauffman@state.or.us or from the Coos County Health Department at (541) 756-2020. Information is also available on the worldwide web at http://www.oregon.gov/DHS/ph/envtox/mafact.shtml

## Literature Cited


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**Electronic Appendix I: Aquatic Analysts, Inc Phytoplankton Reports (attached electronically)**

**Appendix II GreenWater labs Microcystin Analyses**

GreenWater laboratories — aquatic analysis ... research ... consulting

## Microcystin Analysis Report

**Project: City of Lakeside**
(TLBP)

| Sample Identification | Sample Collection Date |
|---|---|
| 1. N. Ten Mile | 09/12/07 |

**Sample Prep –** The sample was ultra-sonicated to lyse all cells and release toxins. Due to the concentration of the sample, a 2-fold dilution was required to bring the concentration of the sample in range of the standard curve for the MC ELISA.

**Analytical Methodology –** A microcystins enzyme linked immunosorbent assay (ELISA) was utilized for the quantitative and sensitive congener-independent detection of MCs. The current ELISA kit is sensitive down to a detection/quantification limit of 0.15 µg/L. Standard recovery was 104%.

**Summary of Results**

| Sample | MC levels (µg/L) |
|---|---|
| 1. N. Ten Mile | ≈ 5.4 |

ND = not detected above the detection limit
Limit of Detection = 0.15 µg/L

* Microcystin data is contained in accompanying spreadsheet.

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# Microcystin Analysis Report

**Project: TLBP**
(City of Lakeside, OR)

| Sample Identification | Sample Collection Date |
|---|---|
| 1. N. Lake | 09/25/07 |

**Sample Prep –** The sample was ultra-sonicated to lyse all cells and release toxins.

**Analytical Methodology –** A microcystins enzyme linked immunosorbent assay (ELISA) was utilized for the quantitative and sensitive congener-independent detection of MCs. The current ELISA kit is sensitive down to a detection/quantification limit of 0.15 µg/L.

## Summary of Results

| Sample | MC levels (µg/L) |
|---|---|
| 1. N. Lake | ≈ 7.5 |

Limit of Detection = 0.15 µg/L

* Microcystin data is contained in accompanying spreadsheet.

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GreenWater laboratories — aquatic analysis ... research ... consulting

# Microcystin Analysis Report

**Project: TLBP**
(City of Lakeside, OR)

| Sample Identification | Sample Collection Date |
|---|---|
| 1. Tenmile-N. Lake | 10/27/07 |
| 2. Tenmile-N. Lake-Kitchen Tap | 10/27/07 |

**Sample Prep –** The sample was ultra-sonicated to lyse all cells and release toxins.

**Analytical Methodology –** A microcystins enzyme linked immunosorbent assay (ELISA) was utilized for the quantitative and sensitive congener-independent detection of MCs. The current ELISA kit is sensitive down to a detection/quantification limit of 0.15 µg/L.

## Summary of Results

| Sample | MC levels (µg/L) |
|---|---|
| 1. Tenmile-N. Lake | ≈ 0.7 |
| 2. Tenmile-N. Lake-Kitchen Tap | ND |

ND = Not detected above detection limit
Detection/Quantification limit = 0.15 µg/L

* Microcystin data is contained in accompanying spreadsheet

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