

Survey for *Cylindrospermopsis raciborskii*

Final Report

INTRODUCTION:

Cylindrospermopsis Raciborskii is a toxin producing blue-green algae whose ideal breeding grounds are reservoirs, small water storage ponds, and slow-moving rivers. It has been found in various countries around the world, and more recently in the southeastern United States. In 2001 *Cylindrospermopsis* was found blooming and producing toxin in Steuben County, Indiana. It has also been found (but not in bloom condition) in Michigan, Illinois, and Ohio. This blue-green algae does not form a typical scum layer associated with blue-greens. Typically dense algal cells are located in a band several feet from the surface in a lake or reservoir. The cells are small and do not create a color in the water. There is no taste or odor associated with *Cylindrospermopsis* or its toxin.

Through this project, funded by the Midwest Technology Assistance Center, the Environmental Resources Training Center (ERTC) of Southern Illinois University Edwardsville contacted surface water treatment facilities in the state of Illinois advising them about the possibility of this toxin producing algae being present in their surface water source and seeking their assistance in collecting suspect samples. A copy of the letter and instructions for collecting samples is attached at the end of this report. Thirteen samples were collected and returned to ERTC. The treatment and investigation of the samples is described below along with a summary of the results.

METHODS:

Returned water samples containing Lugol's solution were stored under refrigeration. Fifty ml was transferred from each water sample to an appropriately labeled 50 ml graduated cylinder. The contents of each water sample was then allowed to settle for 48 hours. Prior to microscopic examinations 45 ml of water was pipetted out of the cylinder, being careful not to disturb the settled contents in the remaining 5 ml. For each sample, 3 microscope slides were prepared using a nanoplankton counting chamber from Phycotech Inc. The chamber (provided by manufacturer) holds exactly 0.66 ml. Samples were examined for *Cylindrospermopsis raciborskii* using a phase contrast microscope. Other organisms were identified when possible and cells per ml were calculated using the instructions provided by Phycotech Inc. for the particular counting chamber.

RESULTS:

Sample 1:

This sample was from Belleville, Illinois. It was collected from the Illinois River Treatment Plant in Peoria, Illinois on November 12, 2003 at 1:30 pm. For 25x field of view, the average number of cells seen was seven. Cell concentration was calculated as 1,010 cells/ml. Organisms identified in the sample were *Scenedesmus*, *Closterium*, and *Schroederia*. Six unidentified diatom species were seen and eight other species were seen but not identified.

Sample 2:

This sample was from Belleville, Illinois. It was collected from the Ohio River (4100 Ohio) on November 5, 2003 at 1:00 pm. For 25x field of view, the average number of cells seen was seven. Cell concentration was calculated as 1,010 cells/ml. Organisms identified in the sample were *Scenedesmus*, *Pediastrum*, *Actinastrum*, *Schroederia*, and *Closterium*. Nine unidentified diatom species were seen and ten other species were seen but not identified.

Sample 3:

This sample was from New Athens, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was forty. Cell concentration was calculated as 5,772 cells/ml. Organisms identified in the sample were *Merismopedia*, *Actinastrum*, *Stephanodiscus*, *Anabaena*, *Schroederia*, *Closterium*, *Pediastrum*, and *Scenedesmus*. Three unidentified diatom species were seen and nine other species were seen but not identified. Additionally a filiform spiral cyanobacterium was seen but also not identified.

Sample 4:

This sample was from Wilmette, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was zero to one. Cell concentration was calculated using 0.5 cell average and was found to be 72 cells/ml. There were only two organisms noted and both were not identifiable.

Sample 5:

This sample was from Waverly, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was twenty-three. Cell concentration was calculated as 3,319 cells/ml. Organisms identified in the sample were *Merismopedia*, *Scenedesmus*, *Actinastrum*, *Closterium*, *Pediastrum*, and *Tetraedron*. One unidentified centric diatom was seen. Twenty-one other species were seen but not identified, and an unidentified Filiform spiral cyanobacterium was seen.

Sample 6:

This sample was from Kankakee, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was six. Cell concentration was calculated as 866 cells/ml. Organisms identified in the sample were *Stephanodiscus*, *Scenedesmus*, *Dinobyron*, and *Fragilaria*. Five unidentified diatom species were seen and ten other unidentified species were seen.

Sample 7:

This sample was from Greenfield, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was zero to one. Cell concentration was calculated using 0.5 cell average and was found to be 72 cells/ml. Organisms identified in the sample were *Pediastrum*, *Scenedesmus*, and *Closteridium*. Five other species were seen but not identified.

Sample 8:

This sample was from Winnetka, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was zero to one. Cell concentration was calculated using 0.5 cell average and was found to be 72 cells/ml. Organisms identified were *Scenedesmus* and *Pediatrum*. Two other species were seen but not identified.

Sample 9:

This sample was from Elgin, Illinois. It was collected from the Fox River on October 8, 2003 at 1:40 pm. For 25x field of view, the average number of cells seen was forty-four. Of the total cells counted an average of 4 cells were found to be *Cylindrospermopsis raciborskii*. Cell concentration was calculated as 6,349 cells/ml. Cell concentration of *C. raciborskii* per ml was calculated as 577 cells/ml. Organisms identified were *Pediastrum*, *Hydrodictyon*, *Ceratium*, *Tetraedron*, *Anabaena*, *Scenedesmus*, *Closterium*, *Treubaria*, *Tribonema*, *Actinastrum*, and *Cylindrospermopsis raciborskii*. Four unidentified diatom species were seen and seven other species were seen but not identified.

Measurements and microscopic examination of *Cylindrospermopsis raciborskii* were as follows: Morphology of *C. raciborskii*: Trichomes solitary, straight, some were blunt at the posterior end, while other trichomes were tapered at the posterior end. Trichomes were seen with both elongated tapered developing heterocysts at the anterior end and developed heterocyst. Trichomes were measured from 124 – 300 μm long by 4 – 6 μm wide (avg. 214.8 μm long by 4.5 μm wide). Vegetative cells are cylindrical with gas vesicles, and measured 10 μm long by 4 – 6 μm wide. Heterocysts seen were terminal at the anterior end, and measured 6 – 30 μm long by 4 μm wide (avg. 13 μm by 4 μm). Akinetes were located next to heterocysts only at anterior end of trichome, and measured 8 – 20 μm long by 4 – 8 μm wide (avg. 16 μm long by 5.9 μm wide).

Sample 10:

This sample was from Blandinsville, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was zero to one. Cell concentration was calculated using 0.5 cell average and was found to be 72 cells/ml. Organisms identified were *Ceratium*, *Pediastrum*, and *Hydrodictyon*. Three other species were seen but not identified.

Sample 11:

This sample was from Salem, Illinois. No collection information was given. For 25x field of view, the average number of cells seen was six. Cell concentration was calculated as 866 cells/ml. Organisms identified were *Anabaena*, *Closterium*, *Actinastrum*, *Merismopedia*, *Tetraedron*, *Pediastrum*, *Staurastrum*, *Treubaria*, *Crucigenia*, and *Scenedesmus*. The sample also contained one unidentified diatom species, one unidentified filamentous cyanobacterium, and seven other unidentified species.

Sample 12:

This sample was from Belleville, Illinois. It was collected from the Mississippi River at Alton, Illinois on September 10, 2003 at 9:30 am. For 25x field of view, the average number of cells seen was thirteen. Cell concentration was calculated as 1,876 cells/ml. Organisms identified were *Asterionella*, *Anabaena*, *Stephanodiscus*, and *Schroederia*. There was also one unidentified diatom species and seven other unidentified species.

Sample 13:

This sample was from Belleville, Illinois. It was collected from a reservoir on December 10, 2003 at 10:00 am. For 25x field of view, the average number of cells seen was twenty-seven. Cell concentration was calculated as 3,896 cells/ml. The sample contained one unidentified diatom species, and six other unidentified species one of which constituted about 99% of cell counts (the cell was oblong, measured 12 x 12 μ m, contained a clear circle at the anterior end with one flagellum, while the rest of the cell appeared medium brown with a black line extending from the bottom of the circle to the posterior end of the cell).

SUMMARY

Thirteen suspect samples from rivers or reservoirs were submitted to ERTC for examination. *Cylindrospermopsis racborskii* was found in one of those samples, which came from the Fox River near Elgin, Illinois.