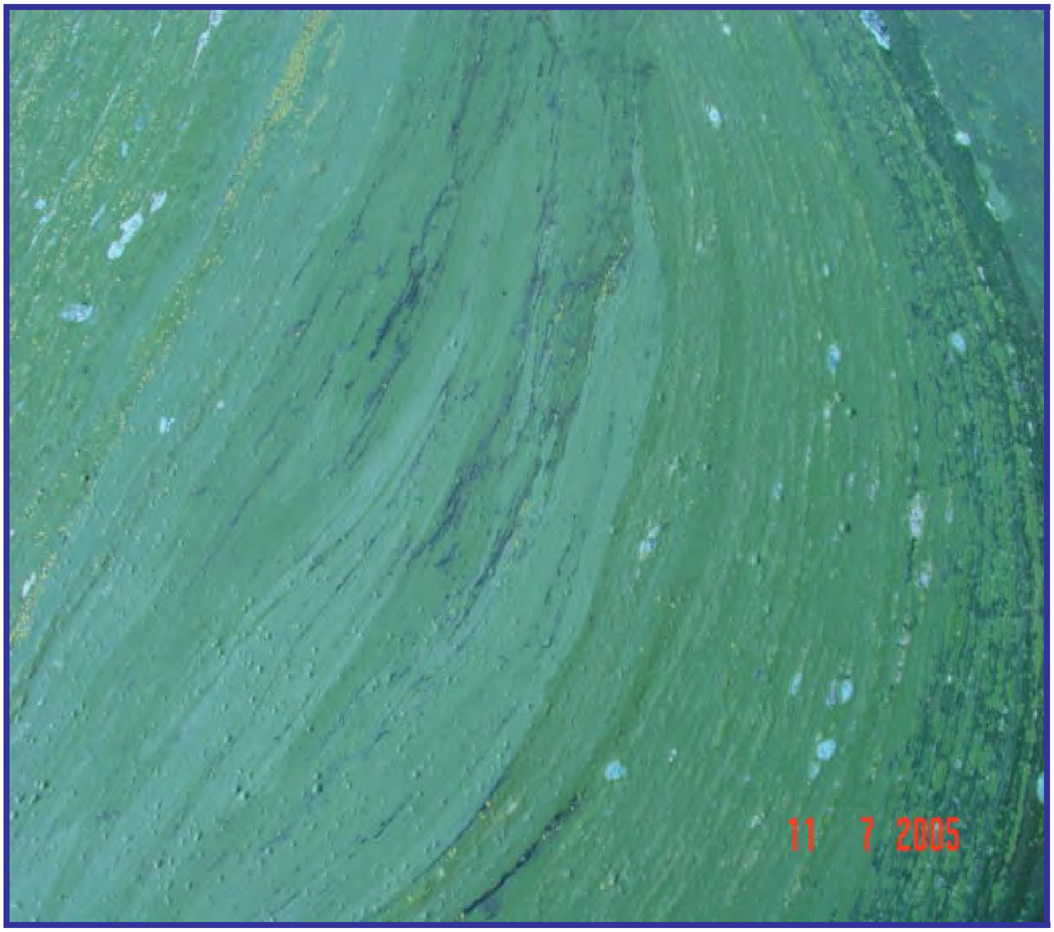
Microcystin Toxin Migration, Bioaccumulation, and Treatment

Fremont Lake #20 Dodge County, Nebraska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Cover Picture: Blue green algae bloom at Wolf Wildcat Reservoir in Gage County, NE.

Introduction

Degradation of water quality in sandpit lakes is a concern in Nebraska. Currently, over 800 publicly and privately owned sandpit lakes exist in the State (NDEQ, 2009). These lakes are typically created from sand and gravel mining operations and are used extensively for recreation by a large, diverse group of people with various interests (i.e. swimming, fishing, SCUBA-diving, hunting). Most sandpits lakes have very small drainage areas and water supplies are via groundwater inputs. A majority of the nutrients found in the water column of a sandpit lake are generated from groundwater and organic matter at the lake bottom. Nutrient loading, particularly phosphorus, has led to accelerated eutrophication of many sandpits throughout the State and has greatly reduced their recreational usage.

Fremont Lake #20 consists of 50 surface acres and is part of a chain of sandpit lakes collectively known as the Fremont State Lakes located near Fremont in Dodge County, Nebraska (Figures 1 and 2). The lake has a maximum depth of 16 foot, average depth of 11 feet, and impounds approximately 552 acre-feet of water (Figure 3). The Nebraska Game and Parks Commission (NGPC) estimates annual usage of the Fremont State Lake Complex to be around 800,000 people.

Lake monitoring conducted in 2005 by the Nebraska Department of Environmental Quality (NDEQ) indicated high concentrations of the microcystin toxin. From 2005 to 2007, approximately 32 percent of the samples exhibited toxin concentrations greater than beach posting target, resulting in a significant loss of water contact recreation opportunities for lake users. Further monitoring conducted from 2005 through 2007 documented high concentrations of phosphorus and nitrogen as being the cause of blue green algae blooms.

The documentation of water quality problems at Fremont Lake #20 resulted in several independent studies being conducted. These studies included an assessment of pre- and post lake treatment nutrient and biological conditions, an evaluation of microcystin toxin accumulation in fish tissue, and an evaluation of microcystin toxin migration out of the lake to groundwater. A significant amount of physical, chemical, and biological data was collected through these studies. This report will summarize the results and conclusions of water quality monitoring efforts at Fremont Lake #20.

Project Partnerships and Funding Sources

Monitoring efforts for the three projects were carried out by several entities. Groundwater monitoring was conducted by the Groundwater Unit, NDEQ. Fish tissue monitoring was conducted by the Surface Water Unit, NDEQ. Pre- and post treatment physical, chemical, and biological monitoring was conducted by the University of Nebraska - Institute of Agriculture and Natural Resources (UNL-IANR), University of Nebraska – Center for Advanced Land Management Technologies (UNL-CALMIT) and the NDEQ Surface Water Unit. Sample analysis was performed by the UNL Water Sciences Laboratory, UNL-IANR Laboratory, Wright State University Laboratory, and NDEQ Laboratory. Monitoring efforts were funded by the University of Nebraska, NDEQ, and U.S. Environmental Protection Agency (USEPA) through Clean Water Act Sections 319 and 106. The Nebraska Environmental Trust (NET) funded the lake treatment and restoration activities. The NGPC was responsible for the alum application and fish renovation.

Figure 1. Location of the Fremont State Lake Complex

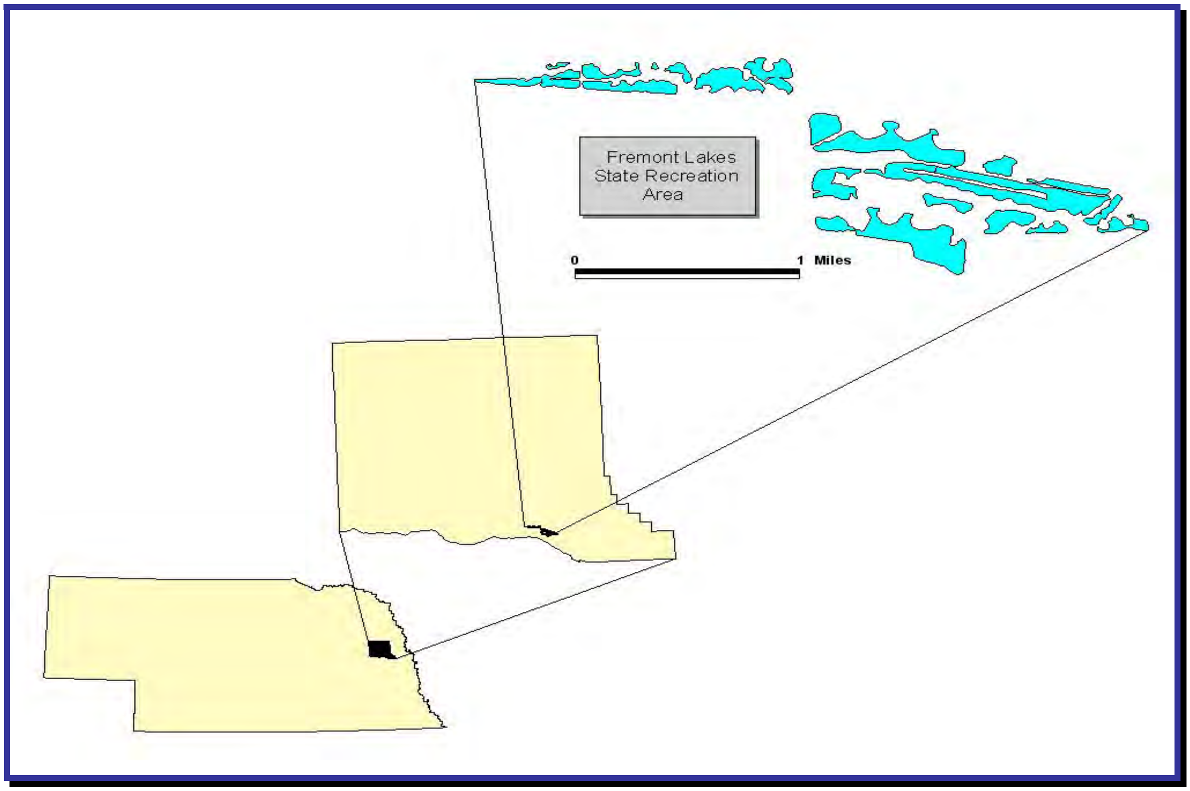


Figure 2. Location of Fremont Lake #20

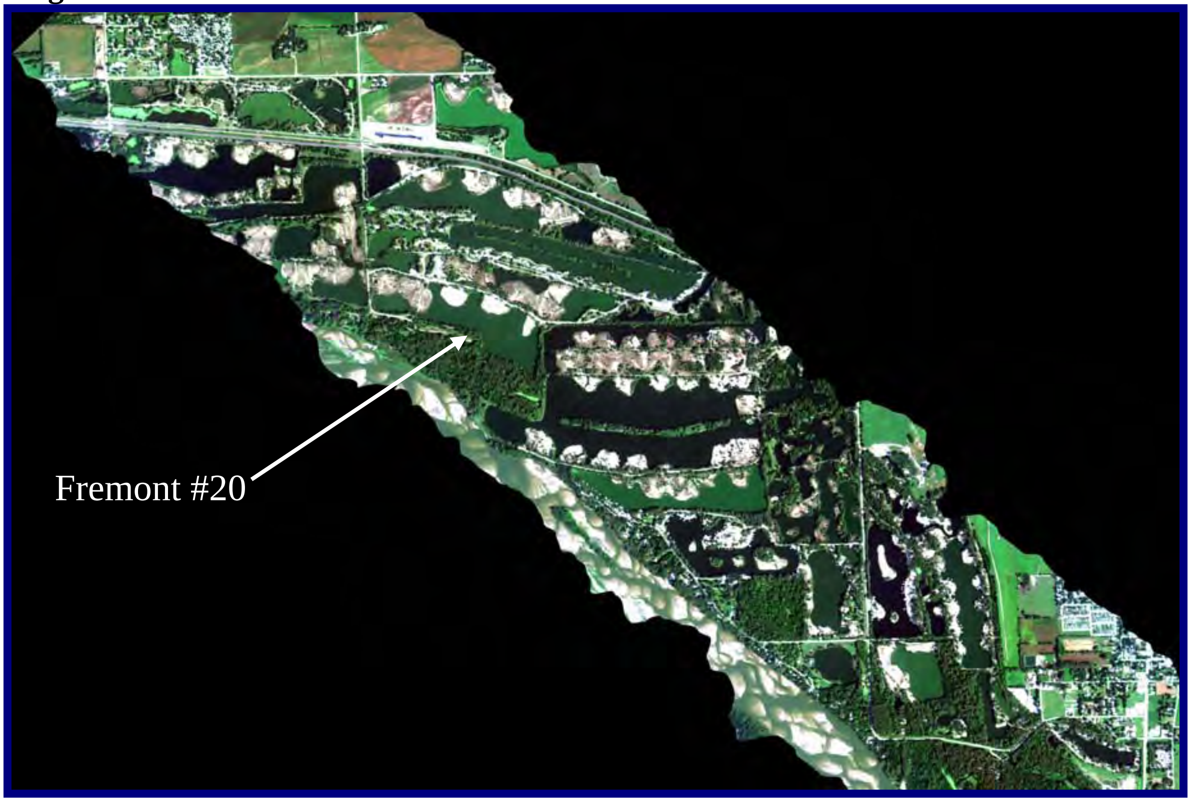
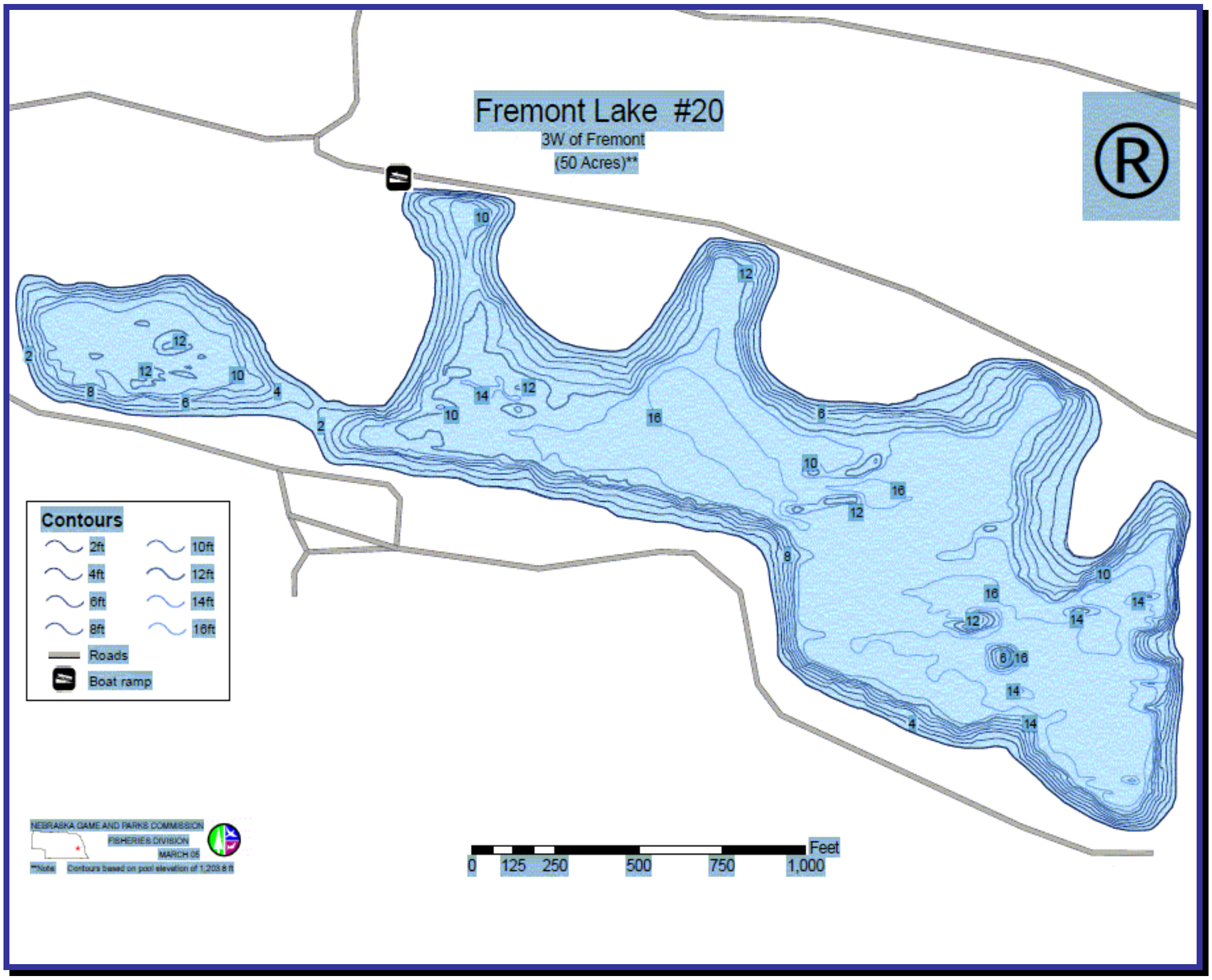


Figure 3. Bathymetric Map of Fremont Lake #20



Water Quality History

It has been well documented that excessive nutrients, particularly phosphorus and nitrogen, are the primary cause of blue green algae blooms (Shindler, 1977). Since Fremont Lake #20 does not have a defined watershed, the only source of nutrients are from groundwater inputs and lake bottom sediments. Studies conducted by UNL and NDEQ indicate organic "muck" on the bottom of the lake as being the major source of internal nutrients. This internal source of nutrients is the result of years of accumulation and decomposition of organic matter (e.g., leaves, algae, dead fish). Internal average phosphorus loads were estimated to be approximately 100 pounds per year based on nutrient analysis of sediment cores; however, this average load could easily be exceeded during extended periods of low dissolved oxygen near the lake bottom.

The state of the fisheries in the lake may have also been a contributor to the problem. The NGPC characterized the fishery as rough fish dominated, primarily with carp and white perch. In addition to direct nutrient contributions through excrement, rough fish can re-suspend nutrients bound to bottom sediments.

In 2008, Fremont Lake #20 was placed on Nebraska's List of Impaired Waters due to impacts to recreation and aquatic life. These impacts were related to excessive nutrient loading and high concentrations of the microcystin toxin.

Lake Treatment Summary

To address blue green algae problems, a nutrient input reduction strategy that targets both external and internal sources needs to be implemented. While external loadings can be addressed through many avenues such as land treatment, fertilizer reductions and waste management, fewer options are available to address internal nutrient loadings.

In the spring of 2006, a project team consisting of representatives from the NGPC, UNL, and NDEQ was established to address blue green algae problems at Fremont Lake #20. The treatment options available to address internal loadings included dredging to remove the organic matter layer at the bottom of the lake, aluminum sulfate treatments to inactivate the phosphorus, and a fisheries renovation.

Hydraulic dredging can be an effective tool to remove organic rich bottom sediments; however, given the size of the lake, hydraulic dredging would not have been cost effective. The cost of removing two feet of organic matter from the bottom of the lake would have been more than $800,000 based on a removal price of $5.00 per cubic yard, which is a very conservative estimate. Aside from the effectiveness being questionable at this lake, additional logistic problems and increased costs with hydraulic dredging existed with the lack of convenient disposal areas. In addition to the cost being high, it would have taken multiple years to hydraulically remove the desired amount of sediment and organic matter. For these reasons, hydraulic dredging was not considered a feasible treatment option.

Alum Treatment

Phosphorus precipitation/inactivation was selected by the project team as the primary treatment option for Fremont Lake #20. This procedure targets the removal of phosphorus from the water column and controls its release from bottom sediments in order to achieve phosphorus limiting conditions to algal growth. The salts of iron, aluminum, and other metals have long been used in advanced wastewater treatment to remove phosphorus and this technique has been extended to lake management. Since early lake treatments in the 1960s, there have been considerable advances in the knowledge of dose, effectiveness, costs, and side effects.

Fishery Renovation

The renovation of the fisheries was selected by the project team as a secondary treatment option. While it was realized that addressing the poor fish community would not sufficiently reduce internal nutrient loads, it was expected to enhance the effectiveness of the alum treatments. The fisheries renovation was conducted in April, 2007. Approximately 552 gallons of rotenone was applied by boat to achieve a 3 ppm treatment of 5 percent liquid rotenone. This was based on a lake mean depth of 10.8 feet. Rotenone is an approved pesticide that targets only gill breathing organisms and breaks down naturally within two weeks. Dead fish were removed from the lake by volunteers and agency staff and properly disposed. In June, 2007 the NGPC restocked the lake with bluegill, largemouth bass and channel catfish.

Prior to treating the lake with alum, the project team made the decision to apply an algaecide to the lake to reduce the amount of algae in the water thus making the alum more effective. A copper sulfate treatment was conducted by a contractor one week prior to the alum treatment. From October 15, 2007 through October 19, 2007 approximately 28,442 gallons of aluminum sulfate and 13,234 gallons of sodium aluminate were applied to Fremont Lake #20. The total cost of the alum treatment was approximately $138,000.

Photo showing alum application to Fremont #20 in October 2007

Migration of Microcystin to Shallow Groundwater

Several years of monitoring in Fremont Lake #20 consistently showed elevated concentrations of the microcystin toxin present in surface water. In April of 2006 the NDEQ Groundwater Unit initiated a groundwater investigation near Fremont #20 in an attempt to determine if the toxin was capable of migrating from surface water to groundwater. The criteria for choosing Fremont Lake #20 were its elevated toxins (greater than the NDEQ advisory level of 20 µg/L), shallow depth to groundwater, coarse sediments, and easy access. To begin generating data on the occurrence of algal toxin levels in vulnerable groundwater settings, nine shallow (~1-4 m) monitoring wells were installed near the lake margins (1 up gradient and 8 down gradient) of Fremont Lake #20. The wells were sampled on a monthly basis for one year from November 2006 though October 2007. The samples were analyzed for the cyclic peptides microcystin and nodularin using an Enzyme-Linked Immunosorbent Assay (ELISA). All environmental data collection was performed under an approved Quality Assurance Project Plan.

Photo of groundwater monitoring event at Fremont #20 in January 2007

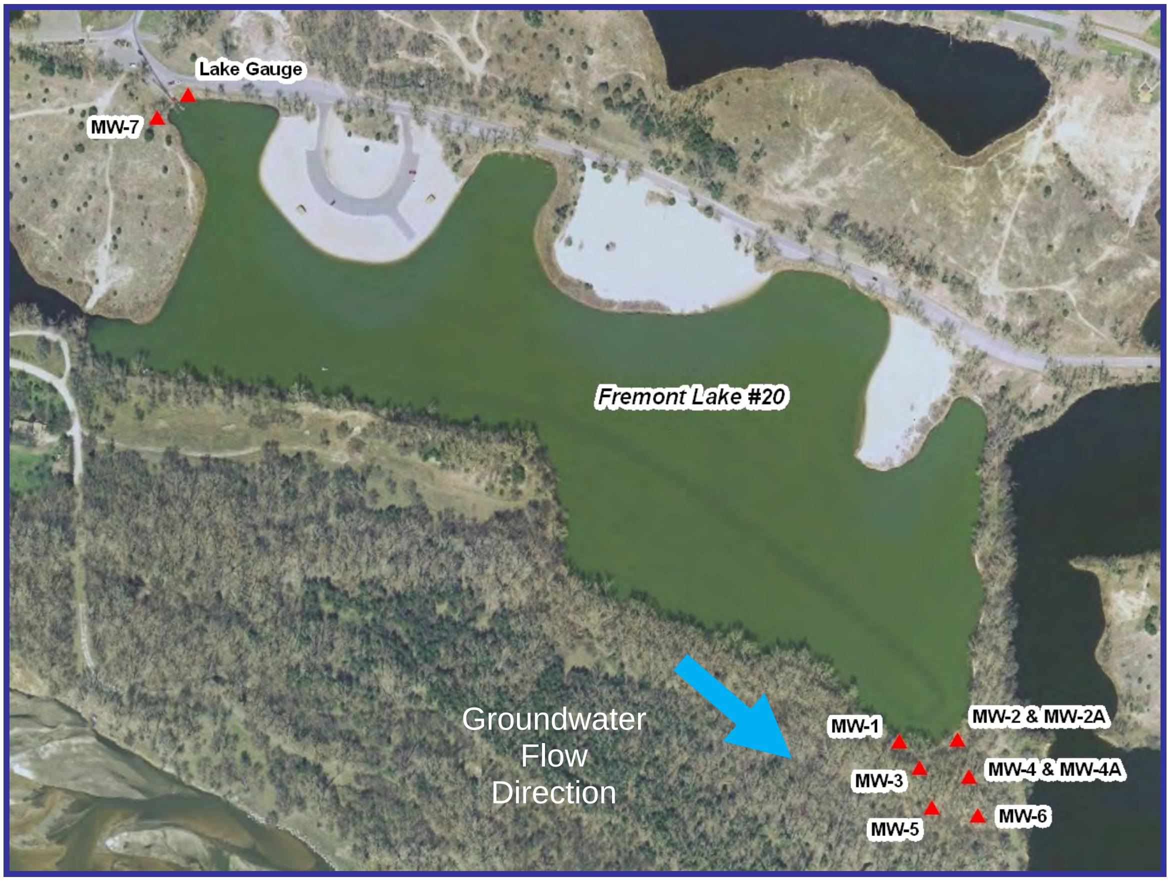
Monitoring Objective

The monitoring objective was to quantify microcystin toxin concentrations in shallow groundwater adjacent to a microcystin impacted surface water body.

Groundwater Concentrations

Groundwater flow direction (east-southeast) and gradient (0.001 ft/ft) were determined using the University of Nebraska-Lincoln Conservation and Survey Division (UNL-CSD) Groundwater flow map 1995. Using an estimated horizontal velocity and gradient, it was calculated that within a month's time the groundwater would migrate 15 feet down gradient. The first set of monitoring wells (MW-1 and MW-2), closest to the shoreline, were placed on the southeast corner of Fremont #20 within fifteen feet of the shoreline. The next two wells (MW-3 and MW-4) were located in areas that were in the down gradient direction (southeast) approximately 90 feet from MW-1 and MW-2. These were followed by a third set of monitoring wells (MW-5 and MW-6) installed approximately 90 feet down gradient (southeast) from MW-3 and MW-4. An up gradient monitoring well (MW-7) on the opposite side of the lake was installed on the northwest corner of Fremont Lake #20 approximately 50 feet from the shoreline. A map showing the locations of the monitoring wells can be found in Figure 4.

Figure 4. Groundwater Monitoring Locations at Fremont Lake #20



A surface water sample was collected from the lake during every groundwater sampling event. A sampling location on the south side of the lake closer to the monitoring well network was chosen. It was decided several months after the groundwater sampling had begun that the surface water samples being collected for the groundwater study were not representative of what was being studied in the groundwater. The toxin would be the only portion that had the potential to migrate through the sediments to groundwater. Collecting raw surface water from the lake had the potential to capture the algae as well. Since the laboratory analyses for microcystin involves freezing the sample to burst the algae cells which then release the toxin, it was determined that the raw surface water would give a false positive. Therefore, two surface water samples were collected each time starting with the November 20, 2006 sampling event. One was filtered using a 0.45µ filter (filtered lake water), and the other one was collected using approved methods (raw lake water).

Only one of the filtered surface water sample (collected 5/21/07) exceeded the NDEQ advisory level of 20 µg/L (Tables 1 and 2). The water quality 30 days prior to this sampling event is unknown since the prior sample was collected on 4/9/07. However, groundwater collected from MW-2 (15 feet from the shoreline) already had exceeded the maximum drinking water concentration of 1 µg/L recommended by the World Health Organization (WHO). The groundwater at the MW-2 location will exceed 1 µg/L for the next two months and the groundwater quality in the wells down gradient from MW-2 (MW-4 and MW-6) were also impacted.

Table 1. Microcystin Concentrations in surface/groundwater (MW-1, 3 & 5)

Date	Up Gradient (MW-7)	Raw lake water	Filtered lake water	Down Gradient (MW-1)	Down Gradient (MW-3)	Down Gradient (MW-5)
11/20/06	No Data	21.48	0.28	<DL	<DL	<DL
12/18/06	No Data	21.09	2.13	<DL	<DL	<DL
1/22/07	<DL	7.11	1.67	1.25	0.18	<DL
2/12/07	<DL	15.80	1.37	<DL	<DL	<DL
3/16/07	<DL	25.60	1.23	<DL	<DL	<DL
4/9/07	<DL	21.48	0.79	0.75	1.00	0.19
5/21/07	<DL	95.69	24.29	0.31	0.16	<DL
6/11/07	<DL	10.86	3.76	0.52	0.40	0.26
7/16/07	0.88	12.80	3.42	0.68	0.27	<DL
8/13/07	<DL	2.65	1.00	<DL	<DL	<DL
9/17/07	<DL	2.57	0.31	<DL	<DL	<DL
10/22/07	<DL	5.52	4.61	<DL	<DL	0.28

DL – Detection Limit

Table 2. Microcystin Concentrations in surface/groundwater (MW-2, 4 & 6)

Date	Up Gradient (MW-7)	Raw lake water	Filtered lake water	Down Gradient (MW-2)	Down Gradient (MW-4)	Down Gradient (MW-6)
11/20/06	No Data	21.48	0.28	<DL	<DL	<DL
12/18/06	No Data	21.09	2.13	<DL	<DL	<DL
1/22/07	<DL	7.11	1.67	<DL	<DL	0.18
2/12/07	<DL	15.80	1.37	<DL	<DL	<DL
3/16/07	<DL	25.60	1.23	<DL	<DL	<DL
4/9/07	<DL	21.48	0.79	0.92	0.63	<DL
5/21/07	<DL	95.69	24.29	4.28	0.95	0.15
6/11/07	<DL	10.86	3.76	6.52	5.49	0.18
7/16/07	0.88	12.80	3.42	1.24	2.42	0.76
8/13/07	<DL	2.65	1.00	0.22	0.25	0.25
9/17/07	<DL	2.57	0.31	<DL	<DL	<DL
10/22/07	<DL	5.52	4.61	<DL	0.17	0.23

DL- Detection Limit

Human Health Risk

The WHO has recommended a maximum drinking water concentration of 1 µg/L microcystin. Most individuals and communities in Nebraska obtain their drinking water from groundwater sources. Results of the sampling indicate that low levels of the toxin can be present in the shallow groundwater surrounding a lake with elevated toxin levels.

Conclusions

If there were a groundwater user within approximately 200 feet down gradient from Fremont Lake #20 when toxin levels exceeded 20 µg/L (in filtered lake water), there would be a possibility that the groundwater quality would be above 1 µg/L for microcystin.

Assessment of Microcystin Toxins in Fish Tissue

In 2005, fish muscle tissue and liver samples were collected from three microcystin impacted waterbodies in Nebraska; Fremont Lake #20 in Dodge County, Carter Lake in Douglas County, and Pawnee Reservoir in Lancaster County. Fish muscle tissue and liver samples were analyzed for the microcystin toxin. All samples were analyzed using an Enzyme-Linked Immunosorbent Assay (ELISA) for the cyclic peptides microcystin and nodularin. All environmental data collection was performed under an approved Quality Assurance Project Plan.

Monitoring Objective

The monitoring objective was to quantify microcystin toxin concentrations in fish fillets and organs to evaluate health risks associated with human consumption.

Muscle Tissue and Liver Microcystin Concentrations

Three species of fish were targeted for collection at Fremont Lake #20; channel catfish, white crappie, and largemouth bass. Two fillet and two liver samples were analyzed from each fish species. One of the two fillet samples and one of the two liver samples from channel catfish exhibited detectable concentrations of microcystin. The detectable concentration from the tissue sample (0.21 µg/g) was comparable to the concentration in the liver sample (0.20 µg/g) (Table 3). All four samples (2 tissue, 2 liver) analyzed from white crappie had concentrations greater than the detection limit (0.147 µg/g). White crappie microcystin concentrations ranged from 0.27 µg/g to 0.32 µg/g. Largemouth bass also had one liver and one tissue sample that exceed the detection limit. Concentrations ranged from 0.21 µg/g to 0.32 µg/g.

While eight of the 12 total samples analyzed from Fremont Lake #20 exhibited detectable concentrations of the microcystin toxin, no samples from the other two lakes exceeded the detection limit (Table 3). A total of 16 samples (8 fillet, 8 liver) were taken from carp, channel catfish, bluegill, and largemouth bass at Carter Lake in Douglas County, none of which had detectable concentrations of the toxin. A total of 18 samples (9 fillet, 9 liver) were collected from carp, channel catfish, bluegill, walleye, white bass, and largemouth bass at Pawnee Reservoir in Lancaster County. None of the 18 samples had detectable concentrations of the microcystin toxin.

Table 3. Microcystin Concentrations in Fish Fillet and Liver Samples

Lake Sampled	Fish Species	Fish Weight (lbs)	Tissue Type	Sample Weight (g)	Microcystin Concentration (µg/g)
Fremont #20	Ch. Catfish	3.32	Fillet	1.0224	<DL
Fremont #20	Ch. Catfish	3.32	Liver	1.2564	0.20
Fremont #20	Ch. Catfish	3.70	Fillet	1.4600	0.21
Fremont #20	Ch. Catfish	3.70	Liver	2.0373	<DL
Fremont #20	Wh. Crappie	0.61	Fillet	1.1250	0.32
Fremont #20	Wh. Crappie	0.61	Liver	0.9227	0.27
Fremont #20	Wh. Crappie	0.79	Fillet	2.1086	0.27
Fremont #20	Wh. Crappie	0.79	Liver	1.4783	0.27
Fremont #20	LM Bass	2.49	Fillet	1.4368	0.32
Fremont #20	LM Bass	2.49	Liver	2.4815	<DL
Fremont #20	LM Bass	2.49	Fillet	2.5164	<DL
Fremont #20	LM Bass	2.49	Liver	2.6242	0.21
Carter	Carp	4.44	Fillet	1.3271	<DL
Carter	Carp	4.44	Liver	1.2698	<DL
Carter	Carp	4.12	Fillet	1.1068	<DL
Carter	Carp	4.12	Liver	2.3543	<DL
Carter	Ch. Catfish	1.41	Liver	2.1746	<DL
Carter	Ch. Catfish	1.41	Fillet	1.7489	<DL
Carter	Ch. Catfish	1.38	Liver	1.9814	<DL
Carter	Ch. Catfish	1.38	Fillet	1.5870	<DL
Carter	Bluegill	0.40	Fillet	1.7837	<DL
Carter	Bluegill	0.40	Liver	1.9995	<DL
Carter	Bluegill	0.31	Fillet	1.4522	<DL
Carter	Bluegill	0.31	Liver	1.6828	<DL
Carter	LM Bass	2.63	Fillet	2.7654	<DL
Carter	LM Bass	2.63	Liver	1.8057	<DL
Carter	LM Bass	2.51	Fillet	2.5719	<DL
Carter	LM Bass	2.51	Liver	1.8800	<DL
Pawnee	Carp	6.22	Liver	2.6319	<DL
Pawnee	Carp	6.22	Fillet	2.1428	<DL
Pawnee	Carp	7.04	Fillet	2.3555	<DL
Pawnee	Ch. Catfish	1.69	Fillet	2.0232	<DL
Pawnee	Ch. Catfish	1.94	Fillet	1.4519	<DL
Pawnee	Ch. Catfish	1.94	Liver	2.0702	<DL
Pawnee	Bluegill	0.49	Fillet	1.7435	<DL
Pawnee	Bluegill	0.49	Liver	0.5594	<DL
Pawnee	Walleye	1.89	Fillet	2.1093	<DL
Pawnee	White Bass	1.25	Fillet	2.3340	<DL
Pawnee	LM Bass	2.88	Fillet	2.5141	<DL
Pawnee	LM Bass	2.88	Liver	1.8555	<DL
Pawnee	LM Bass	2.82	Fillet	2.4150	<DL
Pawnee	LM Bass	2.82	Liver	2.6331	<DL
Pawnee	Carp	7.04	Liver	1.8112	<DL
Pawnee	Ch. Catfish	1.69	Liver	2.7458	<DL
Pawnee	Walleye	1.89	Liver	1.7930	<DL
Pawnee	White Bass	1.25	Liver	2.0088	<DL

Human Health Risk

Nebraska has not yet established numeric objectives or reference guidelines for microcystin toxins in fish tissue. The World Health Organization (WHO) has established a residue guideline value of 0.25 mg/kg of microcystin toxins in finfish fillets (Van Buynder et al. 2001). The WHO has also established a provisional Tolerable Daily Intake (TDI) for microcystin-LR toxin in water of 0.04 ug/kg body weight/day (WHO 1999). While the TDI is not directly related to consuming fish, it is the amount of a potentially harmful substance that can be consumed daily, via ingestion, over a lifetime, with negligible risk of adverse health effects.

Nebraska, like most states, utilizes a risk-based assessment (RBA) procedure similar to EPA's Risk Assessment Methodology (USEPA 1994), to determine the potential for adverse health effects from contaminants in fish. This risk-based assessment procedure utilizes standard equations and estimated exposure parameters, such as ingestion rates and exposure durations, to quantify an individual's risk associated with exposure to a contaminant based on epidemiological studies and animal toxicity studies. However, because specific data from these types of toxicity studies are lacking for microcystin, NDEQ believes it would be inappropriate to assess this fish tissue concentration data using the RBA at this time.

Summary and Conclusions

The microcystin toxin was detected in both fish fillet and liver samples from Fremont Lake #20. Detectable concentrations were found in all three fish species sampled. Findings at Fremont Lake #20 were not consistent with findings from two other lakes sampled. Fillet and liver samples collected from Pawnee Reservoir and Carter Lake exhibited no detectable concentrations of microcystin in either fillet or liver samples. Microcystin concentrations in the lake water columns at the time of sampling were not significantly different. These sampling results likely reveal the inherent difficulty in trying to correlate microcystin concentrations in fish tissue to exposure. It is likely that a fish's diet, its ability to move in and out of the bloom area, and the overall size of the waterbody likely influences tissue and organ concentrations.

While concentrations of microcystin observed in the tissue and livers from fish collected from Fremont Lake #20 could pose a human health risk, the overall indication currently found in the literature is that consumption of fish muscle tissue is not to be considered a major hazard to human health (Hudnell 2008).

Alum Treatment Effectiveness Study

Surface water quality monitoring at Fremont Lake #20 was initiated in 2005 and was continued through 2009. Parameters of concern included phosphorus, nitrogen, chlorophyll *a*, microcystin toxins, phycocyanin, pH, dissolved oxygen, dissolved aluminum, water clarity, and plankton community structure. Sample collection was focused on the "growing season" which is defined as April through September. Most parameters were sampled weekly in 2005, 2007, and 2008. Monthly sampling was conducted in 2006 and in 2009 samples were collected bi-weekly. Microcystin toxin sampling was conducted weekly from 2005 through 2009. All parameters, except for microcystin toxins, were monitored at mid-lake Locations 1 and 2 (Figure 5). Microcystin toxins were monitored at beach Locations 3 and 4 (Figure 5). Samples collected in the epilimnion were taken approximately 0.5 meters below the water surface (near surface samples) while hypolimnetic samples were taken at approximately 4.0 meters of depth (near bottom samples). Pre-alum treatment samples consist of years 2005 through 2007 and post alum treatment samples consist of years 2008 through 2009. Sampling locations and methodologies remained consistent throughout the monitoring project. All environmental data collection was performed under an approved Quality Assurance Project Plan.

Monitoring Objective

The primary objective of this monitoring project was to evaluate and document changes in water quality resulting from implemented lake treatments.

Phosphorus

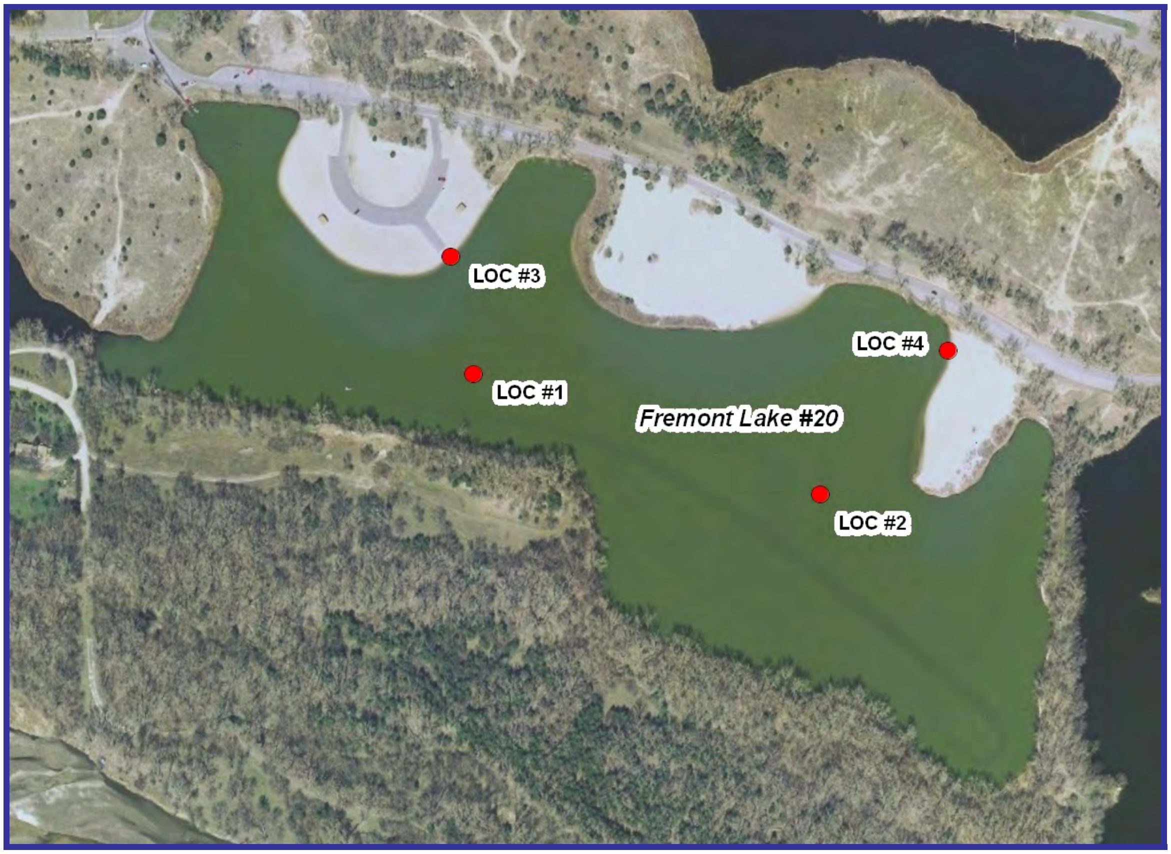
Pre-alum total phosphorus (TP) concentrations near the water surface ranged from 55.7 µg/L in July 2007 to 258.9 µg/L in June 2006 (Figure 6). The pre-alum treatment average TP concentration near the water surface was 127 µg/L.

Post alum treatment TP concentrations near the water surface ranged from 13.6 µg/L in May 2008 to 30.0 µg/L in September 2009. The post alum treatment average TP concentration near the surface was 22 µg/L, which is 83 percent lower than the pre-treatment average.

Pre-alum treatment dissolved orthophosphorus (DOP) concentrations near the surface ranged from 5.6 µg/L in June 2005 to 49.9 µg/L in May 2005. The pre-alum treatment average DOP concentration near the surface was 20 µg/L.

Post alum treatment DOP concentrations near the surface ranged from 5.1 µg/L in September 2009 to 20.4 µg/L in May 2008. The post alum treatment average DOP concentration near the surface was 8 µg/L, which is 59 percent lower than the pre-alum treatment average. Post treatment concentrations of TP and DOP were significantly less (α=.05) than pre-treatment concentrations.

Figure 5. Monitoring Locations at Fremont Lake #20



TP and DOP data were collected near the lake bottom (Hypolimnion) during 2005, 2007, 2008, and 2009. Concentrations of DOP near the lake surface did not reflect internal nutrient release as did the near bottom samples (Figure 7). Several small pulses of DOP were detected in 2005, and in 2007 internal phosphorus loading was significant from early May through July where DOP concentrations jumped from 14 µg/L to 395 µg/L. The internal phosphorus load estimated for May through July, 2007 was 290 pounds. The post alum reduction in DOP near the bottom was approximately 88 percent which is in line with measured reductions of TP in the near surface (84%) and near bottom (87%) samples but was much greater than the reduction calculated for DOP near the surface which was approximately 59 percent.

Peak percentages of DOP in TP samples were observed during late June to early July for all the years assessed (Figure 8). While DOP fractions were generally below 30 percent during 2005, percentages increased to over 60 for most of June, July, and August of 2007. While post alum DOP concentrations were much lower than pre-alum, the percentage of DOP in TP samples remained above 30 percent for most of 2008 and 2009.

Figure 6. Near Surface Total and Dissolved Orthophosphorus in Fremont Lake #20

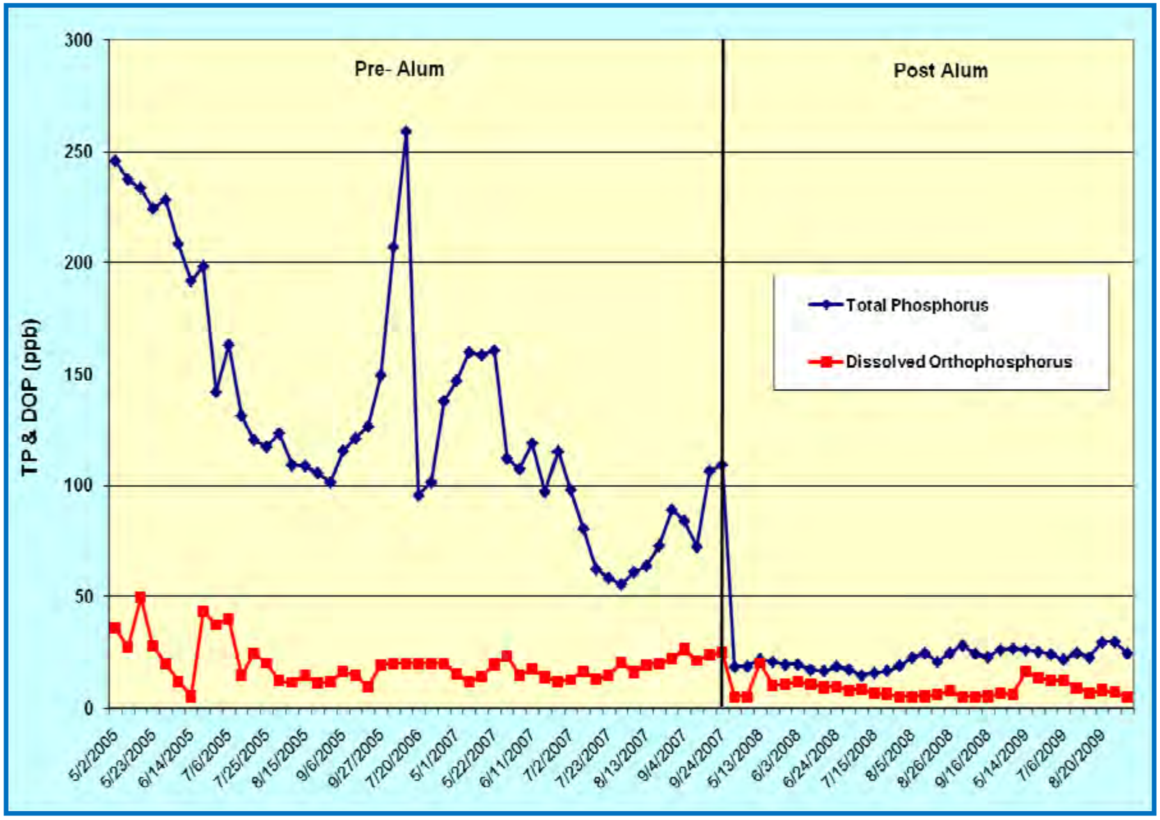


Figure 7. Surface and Bottom Dissolved Orthophosphorus in Fremont Lake #20

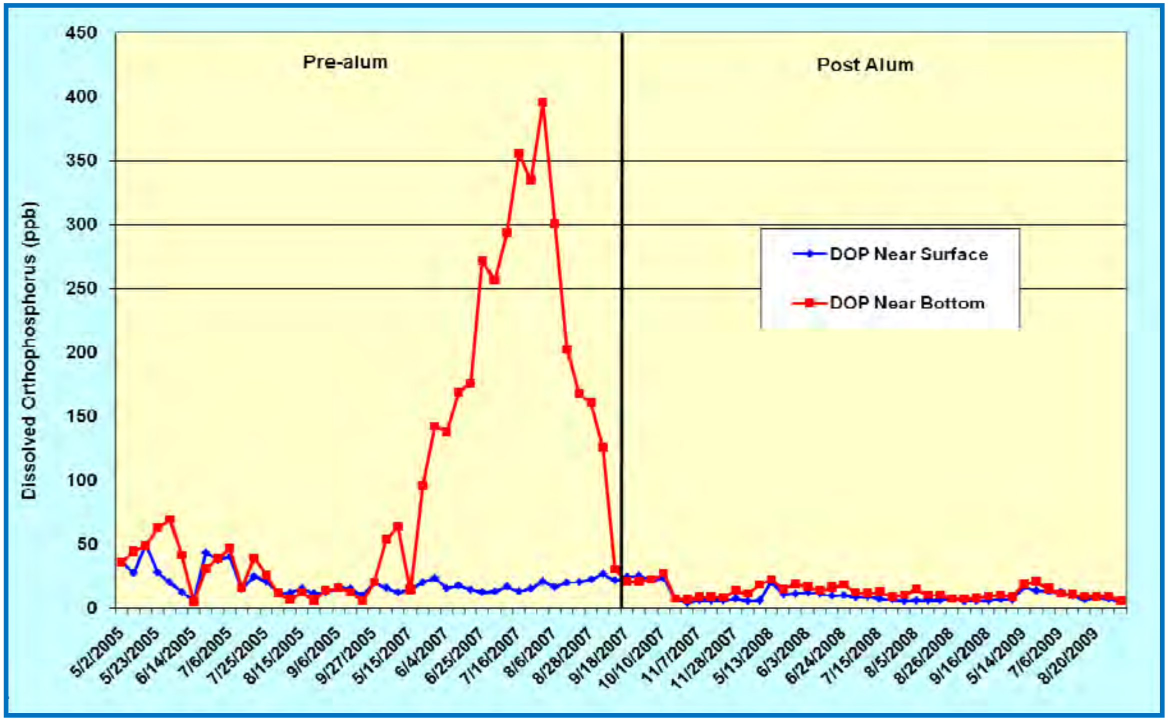
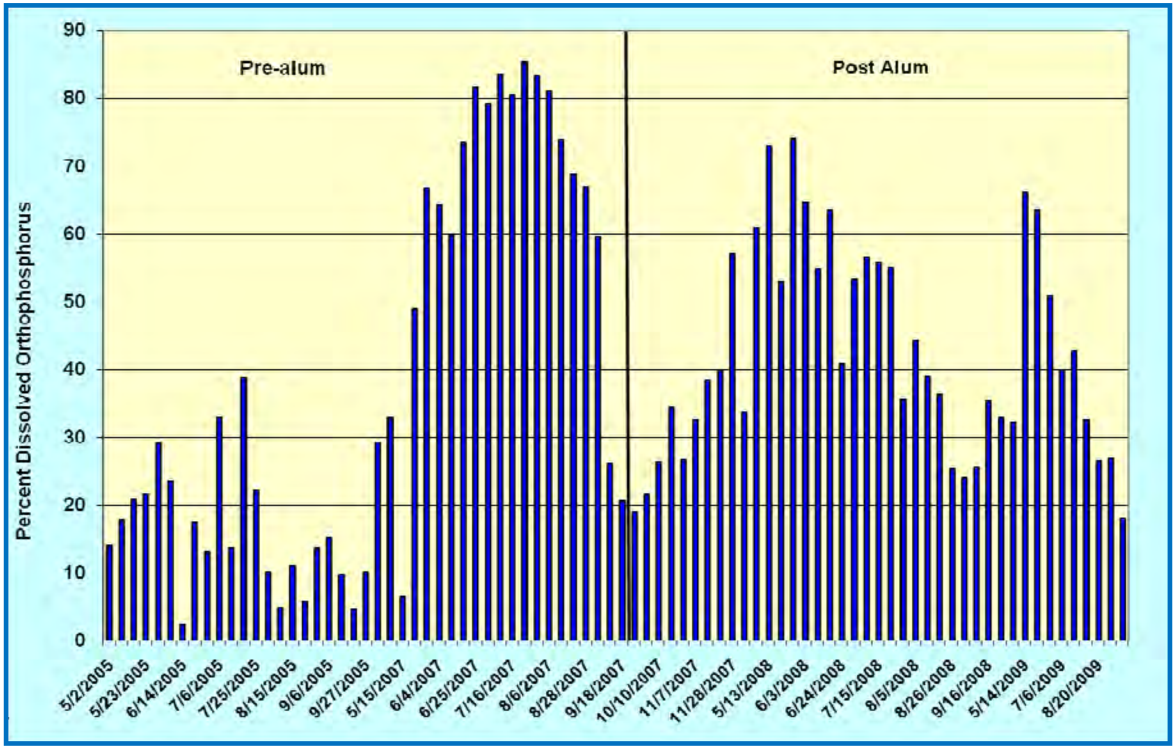


Figure 8. Percent of TP in the Dissolved Fraction in Fremont Lake #20



Nitrogen

Pre-alum total nitrogen (TN) concentrations near the surface ranged from 1,678 µg/L in May 2007 to 4,552 µg/L in August 2005 (Figure 9). The pre-alum treatment average TN concentration near the surface was 2,601 µg/L.

Post alum treatment TN concentrations near the surface ranged from 486 µg/L in June 2008 to 939 µg/L in September 2008. The post alum treatment average TN concentration near the surface was 838 µg/L, which is 68 percent lower than the pre-treatment average. Post treatment TN concentrations were significantly less (α=.05) than pre-treatment concentrations.

While pre-alum concentrations of TN near the surface were significantly different (α=.05) than pre-alum concentrations near the bottom, post alum TN concentrations at the surface were not significantly different than post alum concentrations near the bottom (Figure 10).

The average pre-alum concentration of TN near the bottom was 3,111 µg/L while the post alum concentration near the bottom was 870 µg/L. Post alum concentrations of TN near the bottom were 72 percent less than pre-alum concentrations. Differences in the two data sets were significant (α=0.05).

Figure 9. Near Surface TN in Fremont Lake #20

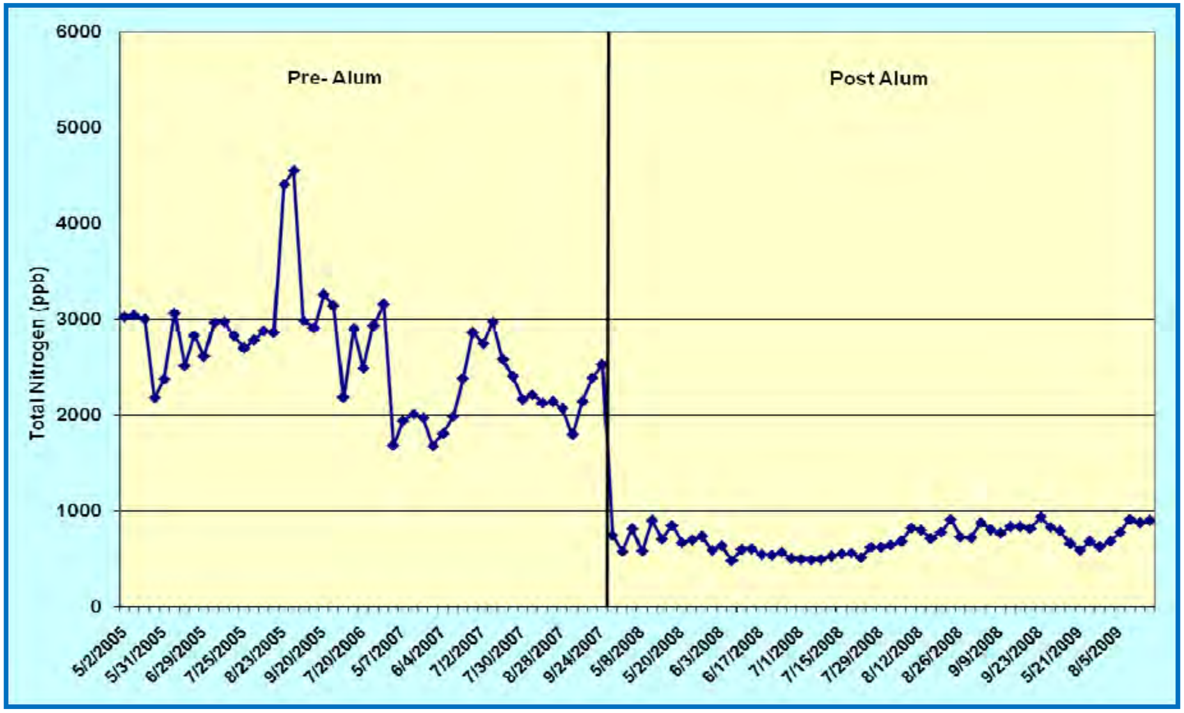


Figure 10. Near Surface and Bottom TN in Fremont Lake #20

Dissolved Oxygen

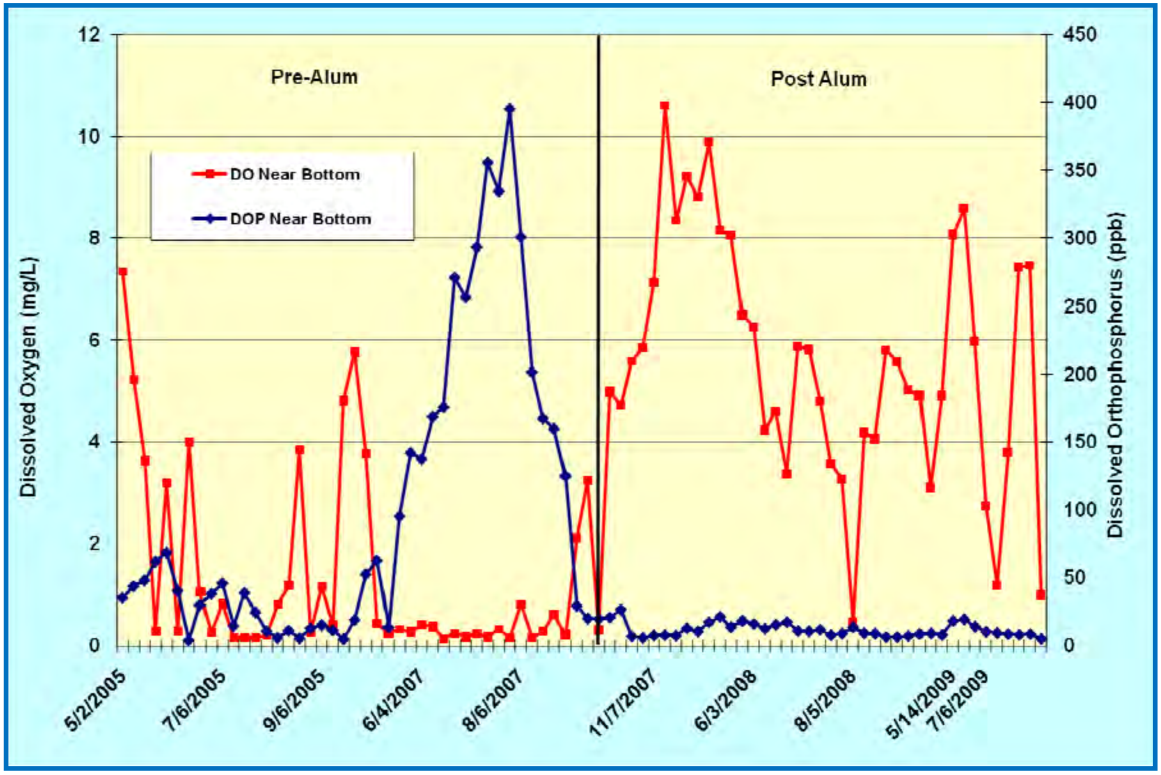
While surface measurements of dissolved oxygen (DO) prior to the alum treatment were not significantly different ($\alpha=0.05$) than measurements taken after the treatment, there were more violations of Nebraska Water Quality Criteria during pre-alum sampling. Nine of 59 measurements (15%) taken prior to the alum treatment were below the standard of 5.0 mg/L while only one of 43 (2%) post treatment measurements were below 5.0 mg/L.

Near bottom measurements of DO fell below 1.0 mg/L on numerous occasions (Figure 11). Pre-alum treatment concentrations were below 1.0 mg/L on 31 of the 59 days sampled or 53 percent of the days sampled. Post alum treatment DO measurements near the lake bottom fell below 1.0 mg/L on only one of 43 days or 2 percent of the days sampled.

Periods of low dissolved oxygen near the lake bottom resulted in phosphorus release from the lake sediments. The period resulting in most internal phosphorus loading was the summer of 2007 where bottom measurements of DO below 1.0 mg/L were measured from May 7th to September 4th (Figure 12). This extended period of hypoxia produced DOP concentrations near the bottom as high as 395 µg/L.

Figure 11. Near Bottom DO in Fremont Lake #20

Figure 12. Near Bottom DO and DOP in Fremont Lake #20



Chlorophyll

Pre-alum chlorophyll *a* concentrations ranged from 5.00 mg/m^3 in September 2006 to 218.08 mg/m^3 in September 2007 (Figure 13). The pre-alum treatment average chlorophyll *a* concentration was 94.66 mg/m^3. Twenty-four of the 49 (49%) pre-alum treatment samples exhibited chlorophyll *a* concentrations greater than 100 mg/m^3.

Post alum chlorophyll *a* concentrations ranged from 2.00 mg/m^3 in June 2008 to 30.74 mg/m^3 in August 2009. The post alum treatment average of 10.22 mg/m^3 was 89 percent lower than the pre-alum treatment average. Post treatment chlorophyll *a* concentrations were significantly less (α=.05) than pre-treatment concentrations.

Water Transparency

Pre-alum water transparency measurements ranged from five inches in September 2006 to 31 inches in August 2007 (Figure 14). The pre-alum average transparency was 16 inches.

Post alum average water transparency measurements ranged from 34 inches in August 2009 to 205 inches in May 2008. The post alum average transparency of 99 inches was 6.2 times greater than the pre-alum average. While post alum water clarity is significantly greater (α=.05) than pre-alum clarity, there has been a decreasing trend since the alum treatment was completed.

Figure 13. Near Surface Chlorophyll *a* in Fremont Lake #20

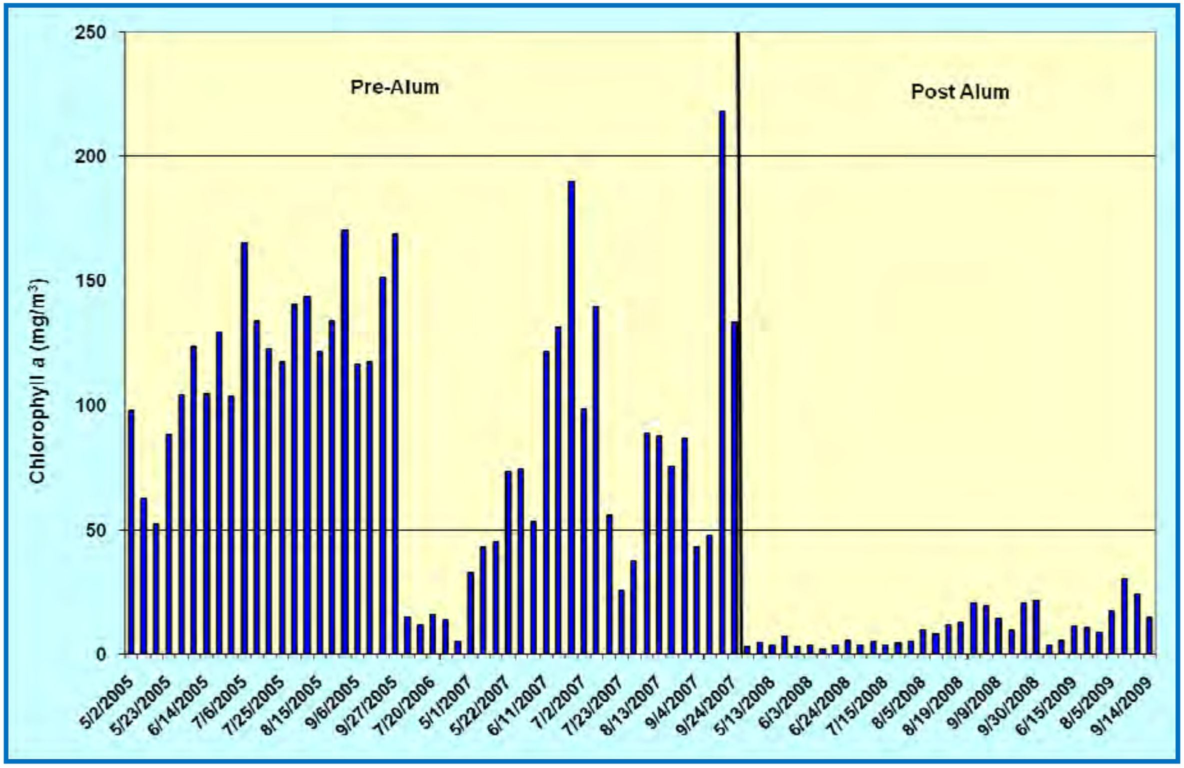
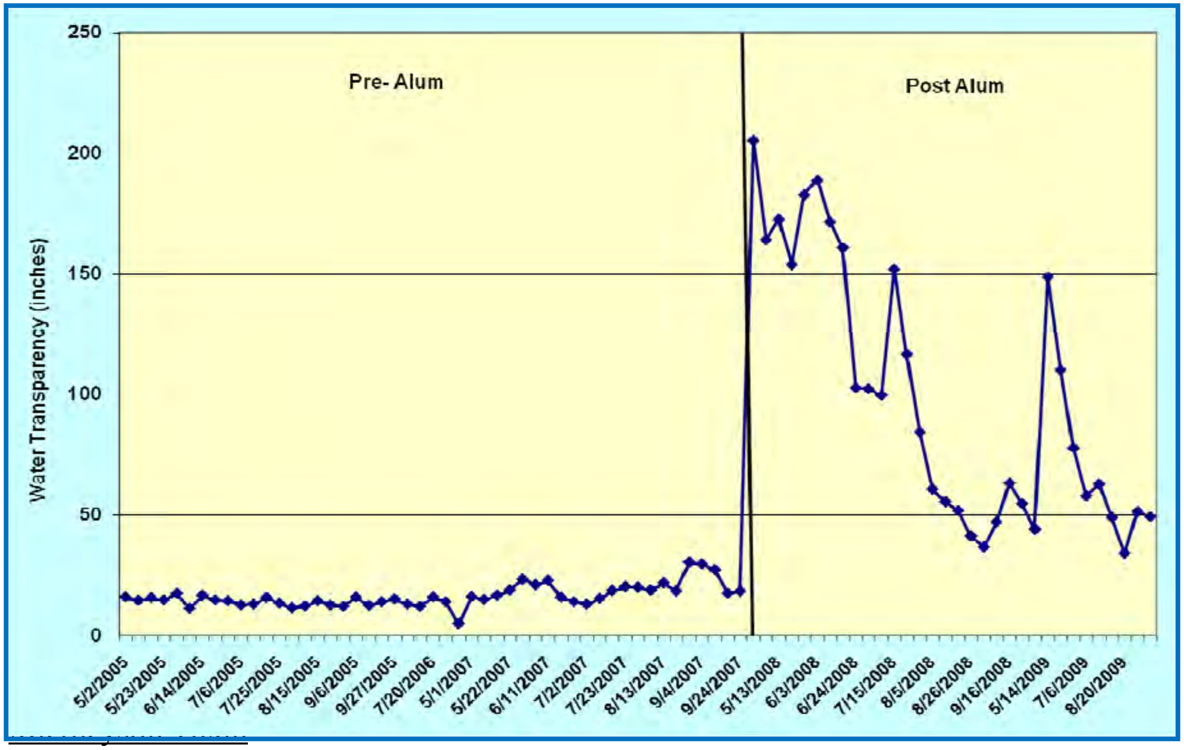


Figure 14. Water Transparency in Fremont Lake #20



Microcystin Toxins

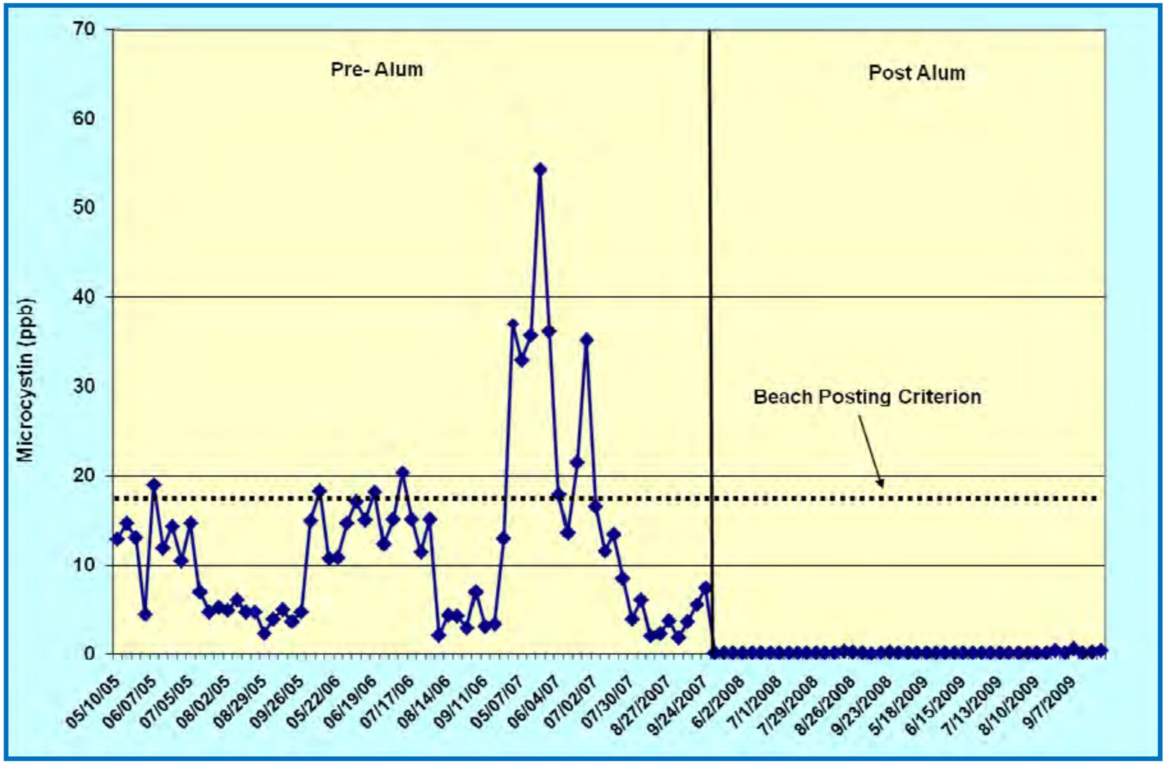
Pre-alum treatment concentrations of the microcystin toxin ranged from the laboratory reporting limit of 0.15 µg/L in August 2005 to 90.16 µg/L in May 2007 (Figure 15). Pre-alum samples exceeded the beach posting criterion of 20 µg/L for 21 of the 65 days sampled. The average microcystin concentration for the pre-alum period was 12.21 µg/L.

Post alum treatment concentrations of the microcystin toxin ranged from the laboratory reporting limit of 0.15 µg/L on numerous occasions to 1.05 µg/L on September 2009. All post alum treatment samples have had concentrations below the beach posting criterion of 20 µg/L. The average post alum treatment microcystin concentration was 0.19 µg/L, which is a 98.4 percent reduction in average concentrations. Post alum concentrations of the microcystin toxin were significantly less (α=.05) than pre-alum concentrations.

Phytoplankton Communities

In 2007, prior to the lake treatments, UNL collected samples for phytoplankton identification and enumeration. Samples were collected at four locations in the lake which remained consistent for all phytoplankton sampling. Integrated phytoplankton samples were collected from the surface down to a depth of two meters using a 2 inch PVC tube fitted with a one-way ball valve. Sample collection, preservation, and processing were completed using methodologies described in the Fremont Lake #20 Water Quality Study QAPP.

Figure 15. Average Microcystin Concentrations in Fremont Lake #20



Pre-alum phytoplankton communities in 2007 were comprised mainly of blue green algae (Figure 16). *Oscillatoria* sp. was the dominant taxa found in the lake, comprising no less than 85 percent of the pre-treatment composition from April through October. In November and December 2007, *Oscillatoria* sp. comprised an average of 69 percent of the total composition. The second most dominant taxa was *Cryptomonas* sp. which comprised an average of one percent of the composition from April through October and averaged 30 percent of the composition in November and December 2007.

Post treatment phytoplankton communities were evaluated in 2008. The number of taxa present increased from 11 during pre-alum to 27 post alum (Table 4). While taxa numbers increased, most taxa constituted a small amount of the total composition. The diatom *Fragilaria* sp. comprised nearly 80 percent of the community in April 2008 (Figure 17). During May the community shifted to green algae *Quadrigula* sp. and *Schroederia* sp. By June, the blue greens *Chroococus* sp. and *Anabaena* sp. comprised 50 percent of the sample and by July *Oscillatoria* sp. comprised 85 percent of the taxa composition.

Figure 16. Pre-Alum Algae Composition in Fremont Lake #20 (2007)

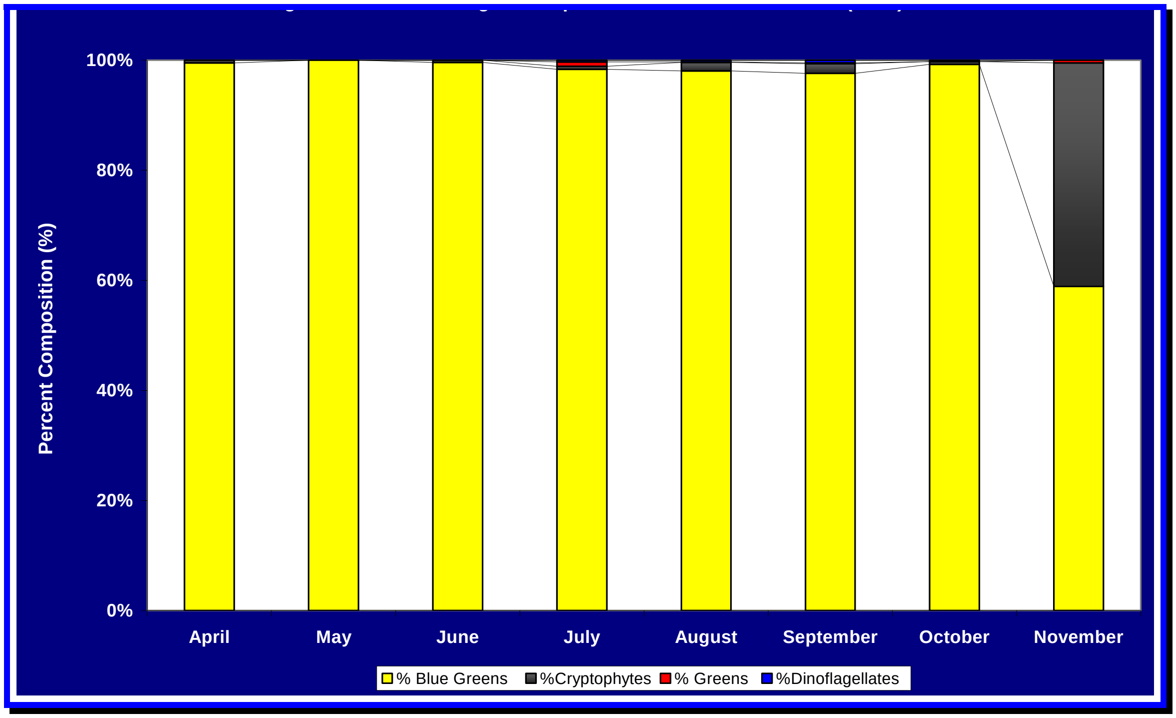
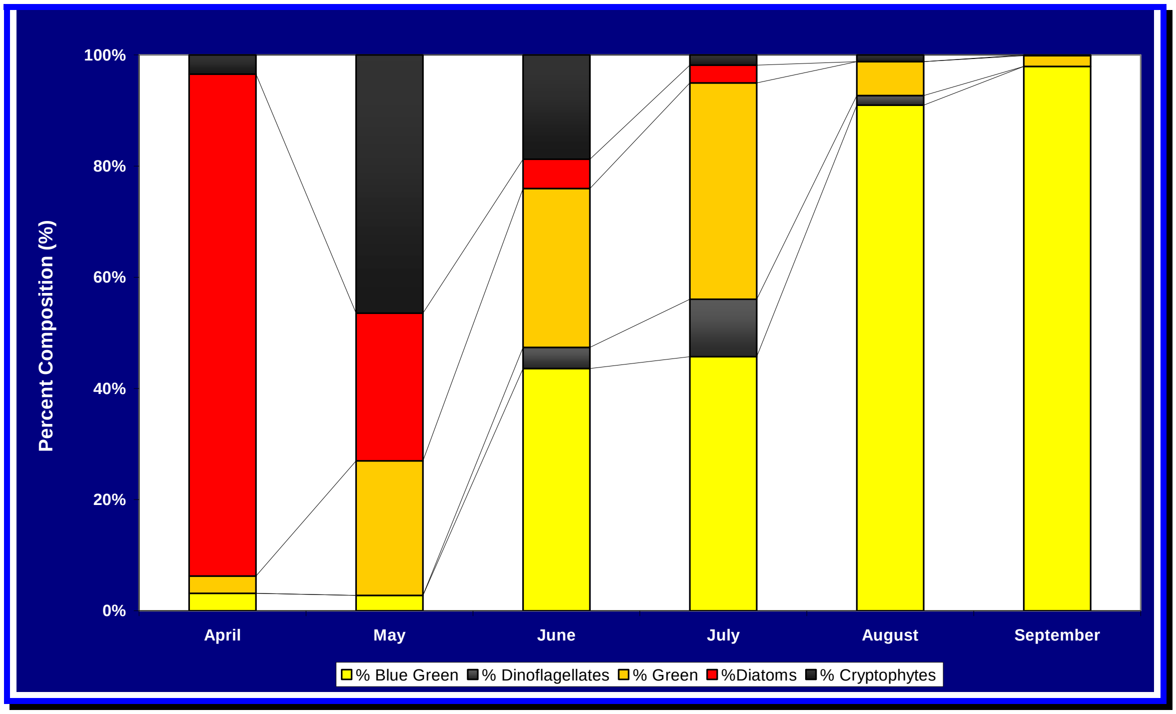


Table 4. Pre- and Post Alum Phytoplankton Taxa List

Taxa	Pre-Alum (2007)	Post Alum (2008)
Oscillatoria sp.	✓	✓
Anabaena sp.	✓	✓
Merismopedia	✓	✓
Chroococcus sp.	✓	✓
Microcystis sp.	✓	✓
Spirulina sp.	✓	✓
Ceratium sp.	✓	✓
Cryptomonas sp.	✓	✓
Pediastrum sp.	✓	✓
Cosmarium sp.	✓	✓
Scendesmus sp.	✓	✓
Ankistrodesmus sp.		✓
Closterium sp.		✓
Desmidium sp.		✓
Quadrigula sp.		✓
Staurastrum sp.		✓
Schroederia sp.		✓
Eudorina sp.		✓
Cymbella sp.		✓
Cyclotella sp.		✓
Pinnularia sp.		✓
Navicula sp.		✓
Asterionella sp.		✓
Fragilaria sp.		✓
Frustulia sp.		✓
Dinobryon sp.		✓
Peridinium sp.		✓

Figure 17. Post Alum Phytoplankton Composition in Fremont Lake #20 (2008)

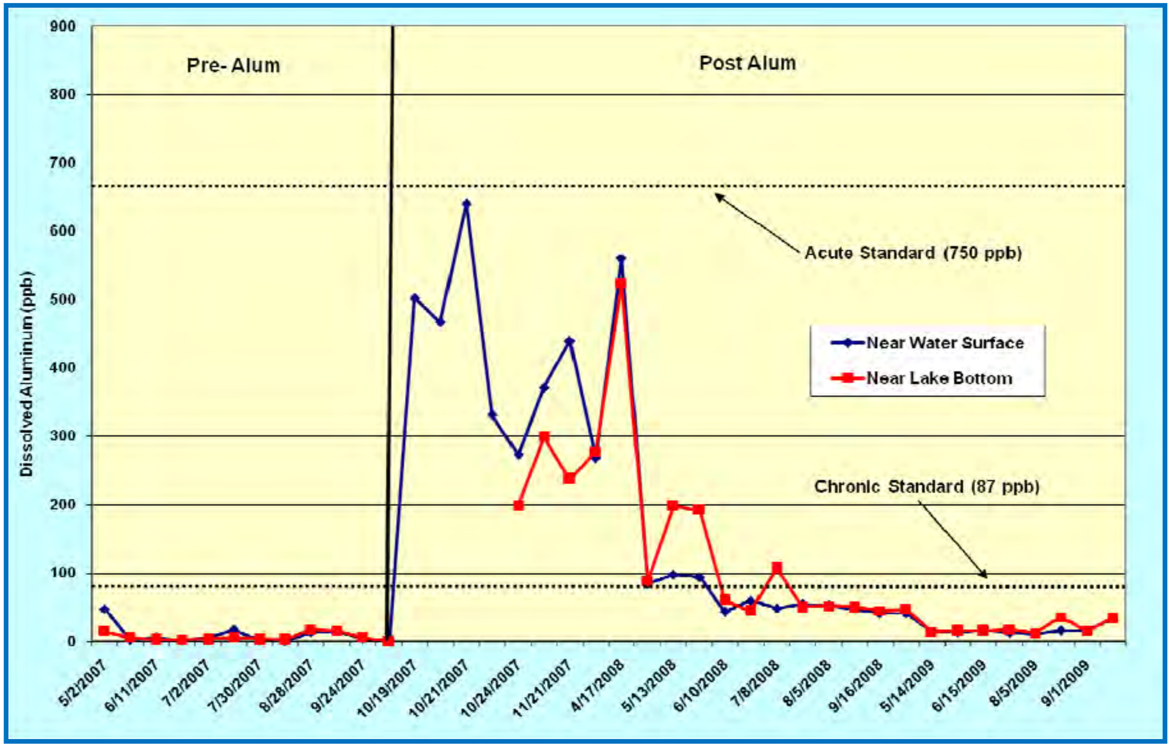


Dissolved Aluminum

Dissolved aluminum was monitored prior to, during, and after the alum treatment. Prior to the treatment, dissolved aluminum concentrations ranged from 1 µg/L in August 2007 to 47 µg/L in May of 2007 (Figure 18). All concentrations were below state chronic and acute water quality standards which are 87 µg/L and 750 µg/L respectively (NDEQ, 2009).

Immediately after the alum treatment, the lake wide average dissolved aluminum concentration increased to 502 µg/L. While concentrations never exceeded acute criteria, they remained above chronic criteria from October 2007 until June of 2008. Since June 2008 concentrations have continued to drop and as of September 2009 concentrations were approaching pre-project levels.

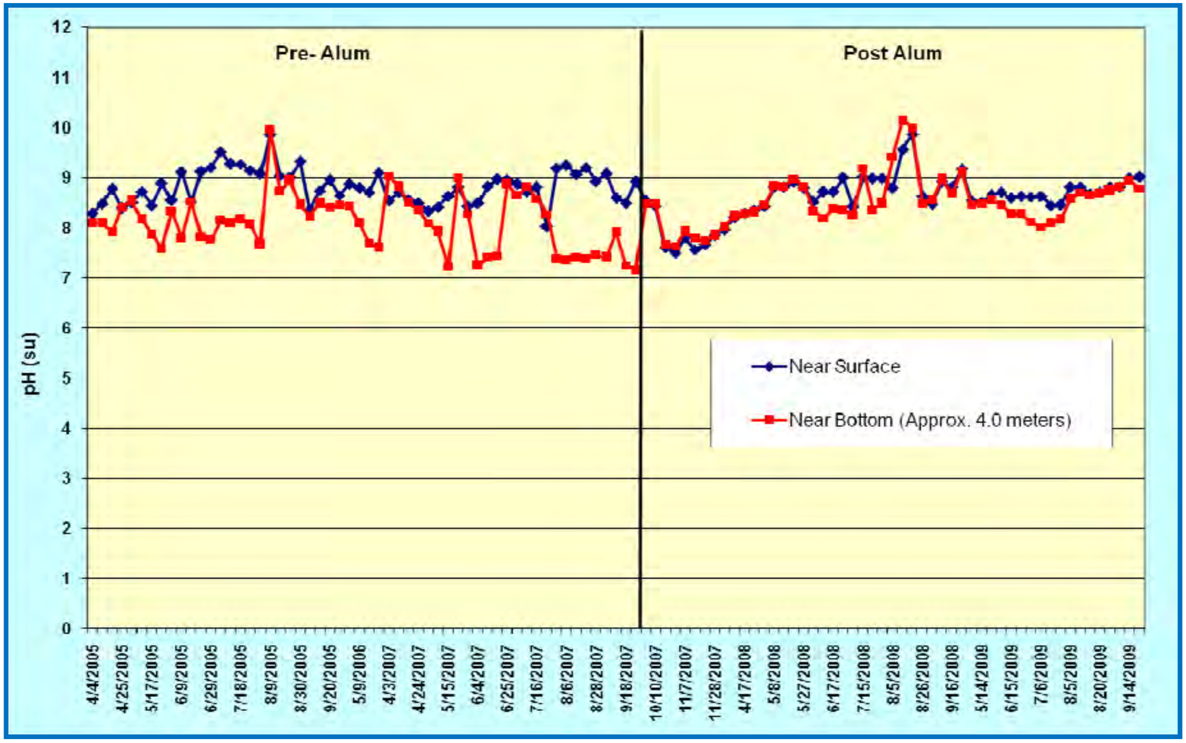
Figure 18. Dissolved Aluminum in Fremont Lake #20



Water pH

Pre-alum pH measurements ranged from 7.16 *su* in September 2007 to 9.97 *su* in August 2005 (Figure 19). The pre-alum average pH was 8.12 *su*. Post alum measurements ranged from 7.63 *su* in October 2007 to 10.15 *su* in August 2008. While surface pH measurements exhibited a slight decrease after the alum treatment, bottom pH measurements exhibited a slight increase. There were no significant shifts in pH during the alum application process.

Figure 19. pH Measurements at Fremont Lake #20



Remote Sensing

Remote sensing techniques are increasingly being used to collect valuable, often unique information about water resources. In general terms, remote sensing involves gathering data and information about the physical "world" by detecting and measuring signals composed of radiation, particles, and fields emanating from objects located beyond the immediate vicinity of the sensor device(s). For Fremont Lake #20, aircraft flyovers were used to remotely sense and record the unique spectral reflectance of chlorophyll (which is a pigment produced by all plant growth in a lake) and phycocyanin (a pigment produced by blue green algae). The spectral reflectance data was then transferred to a map-like image of the lake to illustrate the presence and location of algae growth, and specifically blue-green algae. AISA hyperspectral image data were periodically collected from 2006 through 2008 to evaluate the extent, magnitude, and duration of algae blooms. Pre-alum treatment images were developed for five dates in 2006 and six dates in 2007.

In 2006, chlorophyll densities increased lake wide during a two week period from May 24 to June 6 and densities continued to be high through June 14, 2006 (Figures 20, 21, 22). As chlorophyll concentrations increased during this time period so did the presence of phycocyanin or blue green algae (Figures 23, 24, 25). The increase in blue green algae was also accompanied by a measured increase in the microcystin toxin as concentrations increased from 9.79 µg/L on May 26, 2006 to 17.05 µg/L on June 5, 2006, and continued to increase to 23.87 µg/L on June 12, 2006. While chlorophyll concentrations started to decrease by the time the last flight was conducted on August 23, 2006, a lake wide presence of phycocyanin was still observed and microcystin toxin concentrations were around 4.94 µg/L (Figures 26 and 27).

Images for 2007 showed a similar spring pattern with chlorophyll although lake wide presence of phycocyanin in 2007 didn't occur until early July. Chlorophyll *a* concentrations were 189.60 mg/m3 on June 25, 2007 and 139.84 mg/m^3 on July 9, 2007 but dropped to 87.04 mg/m3 by August 28 (Figures 28, 29, 30). Microcystin toxin concentrations greater than 20 µg/L were reported in May and June but by the end of July 2007, concentrations were down to 3.96 µg/L.

Imagery documented another algal bloom that occurred between September 4th, 2007 and September 26, 2007 (Figures 31, 32, 33, 34). This bloom was also accompanied by an increase in microcystin toxin concentrations. Concentrations increased from 1.86 µg/L on September 4th to 7.41 µg/L on September 24th.

Post alum treatment images were developed for 11 dates in 2008. Images identified low chlorophyll concentrations and low phycocyanin throughout 2008 (Figures 35 through 40). Imagery for 2008 corresponds well to chlorophyll and microcystin toxin data as low concentrations for both have been measured.

As mentioned, one benefit of using remote sensing information is the amount of land area that can be assessed at any given time. Due to these capabilities, imageries produced for Fremont Lake #20 also included data from several surrounding public and private lakes that were developed in the same time period. Blooms observed at these lakes exhibit similar patterns from 2006 through 2008 indicating the change in conditions at Fremont Lake #20 were most likely not due to natural environmental factors. In addition, other lakes identified as a problem based on chlorophyll and phycocyanin imagery are now being targeted for monitoring.

Figure 20. Chlorophyll Remote Sensing Image for Fremont State Lakes

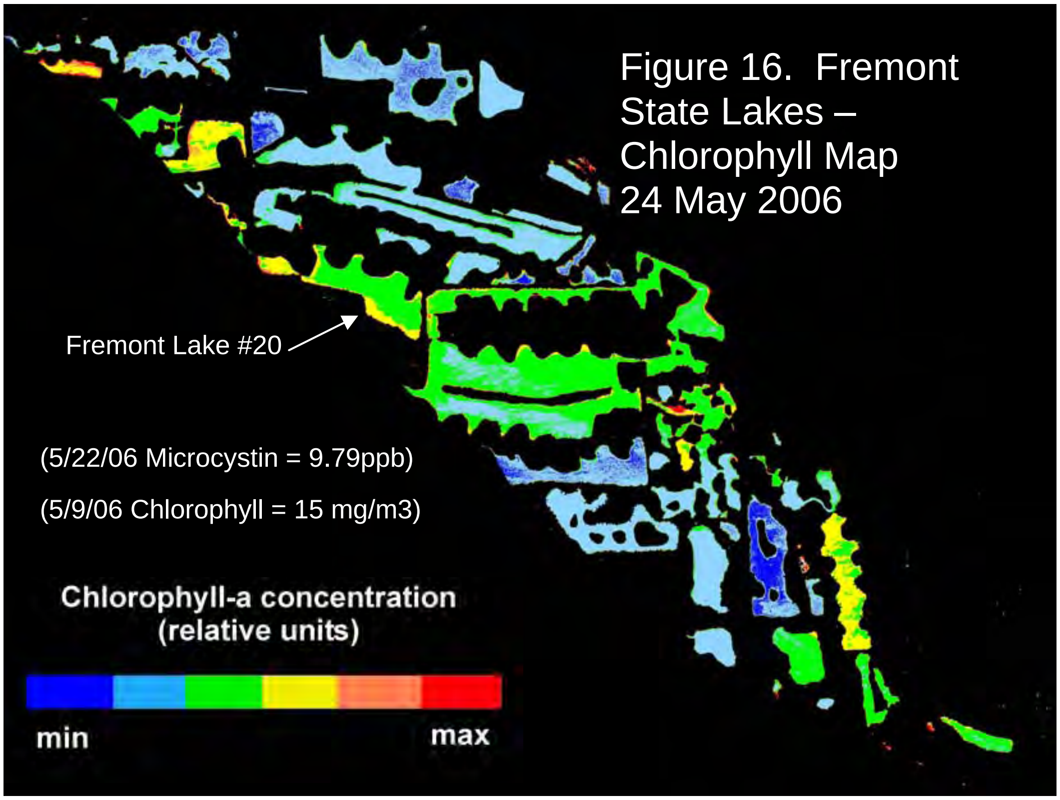


Figure 21. Chlorophyll Remote Sensing Image for Fremont State Lakes

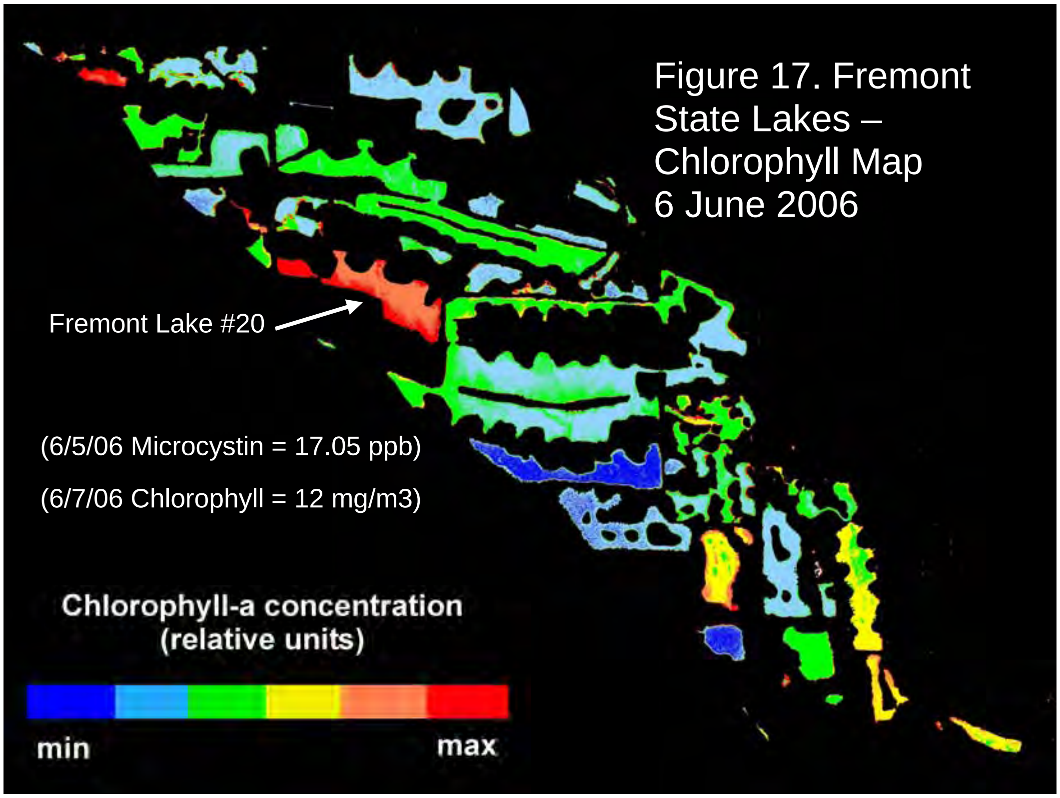


Figure 22. Chlorophyll Remote Sensing Image for Fremont State Lakes

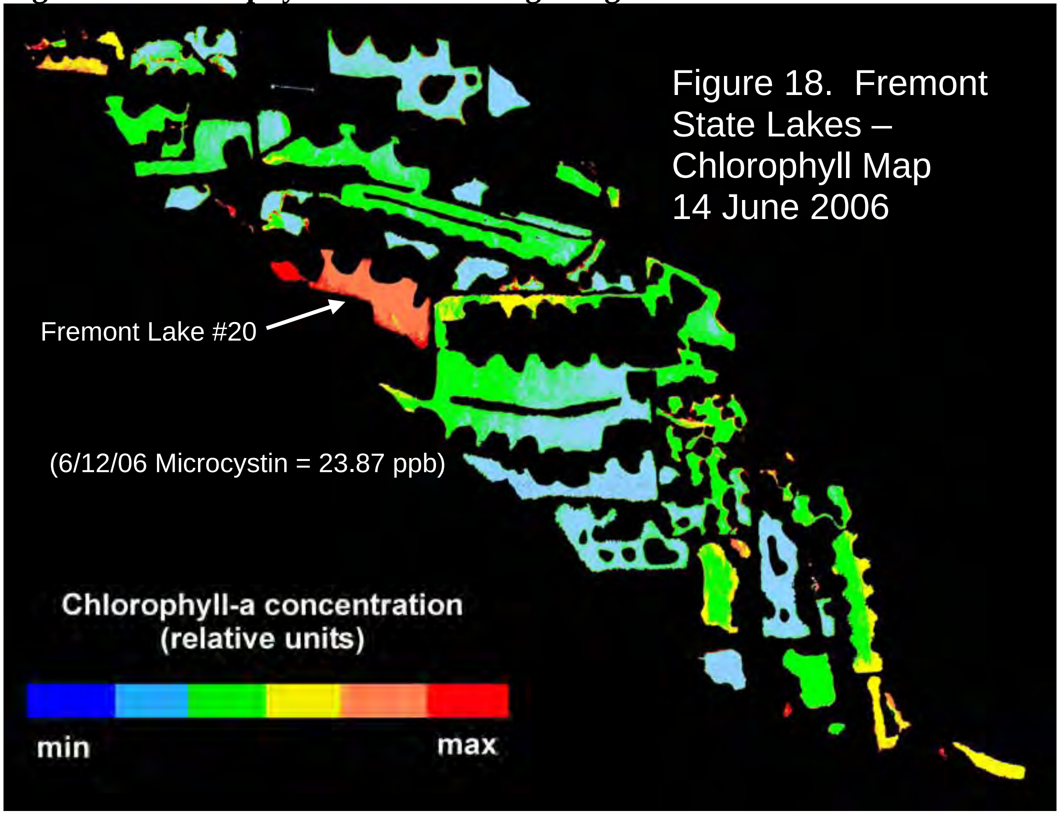


Figure 23. Phycocyanin Remote Sensing Image for Fremont State Lakes

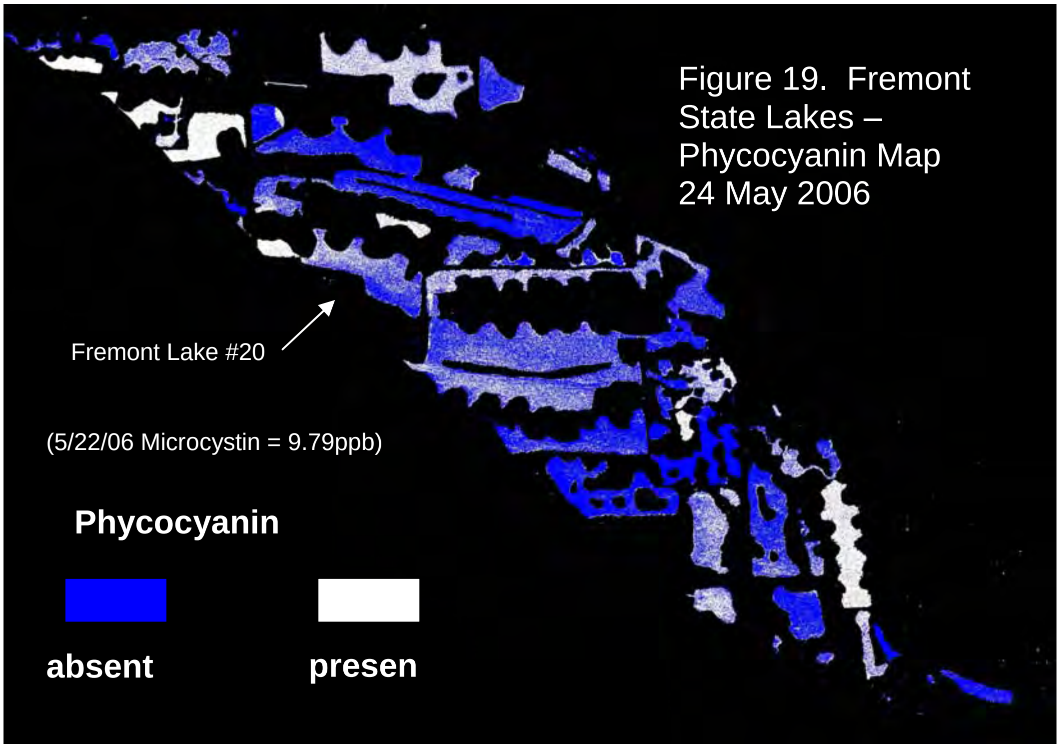


Figure 24. Phycocyanin Remote Sensing Image for Fremont State Lakes

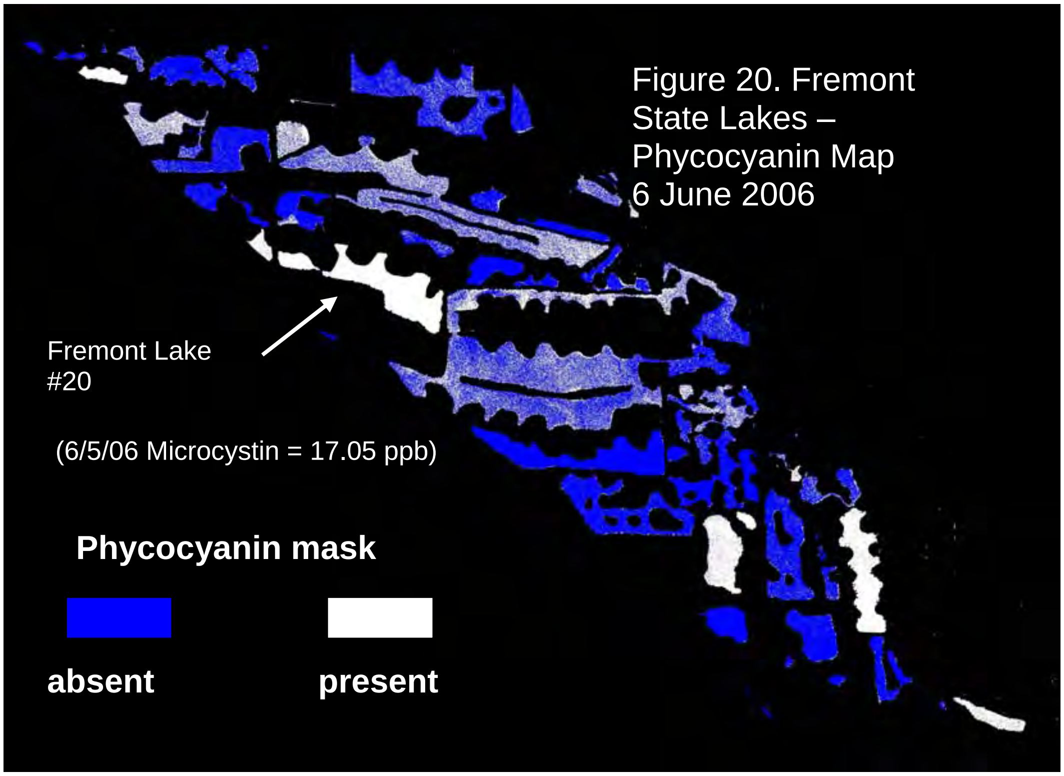


Figure 20. Fremont State Lakes – Phycocyanin Map 6 June 2006

(6/5/06 Microcystin = 17.05 ppb)

Figure 25. Phycocyanin Remote Sensing Image for Fremont State Lakes

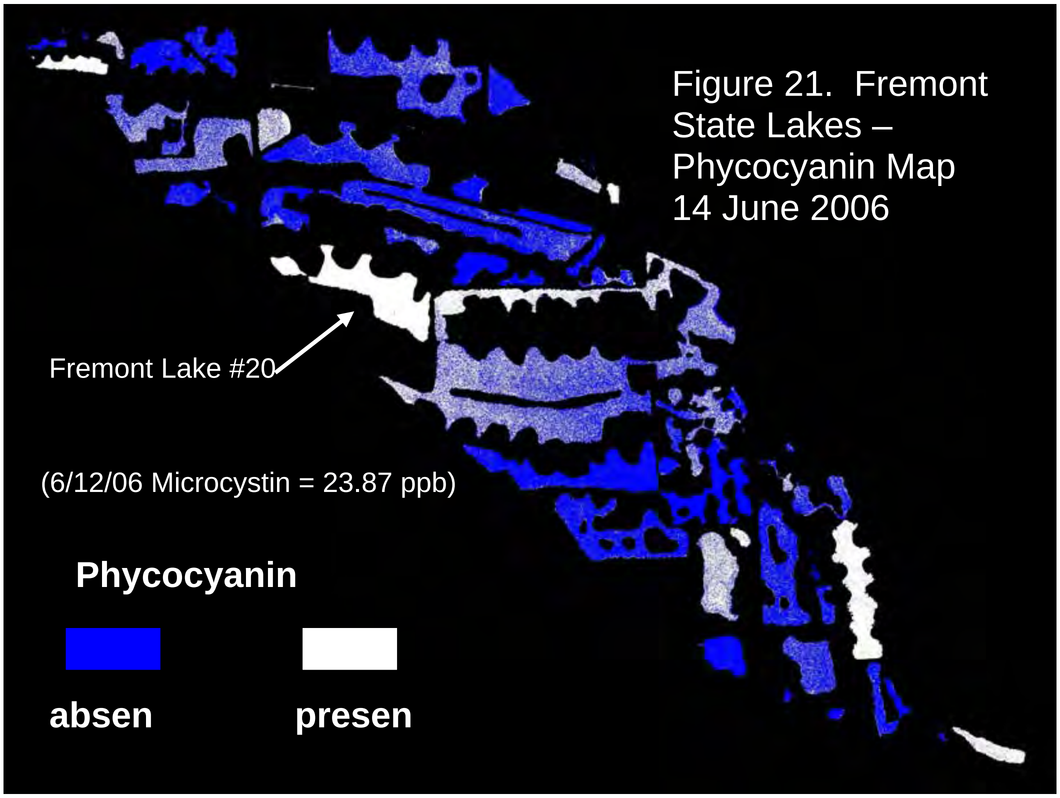


Figure 21. Fremont State Lakes – Phycocyanin Map 14 June 2006

(6/12/06 Microcystin = 23.87 ppb)

Figure 26. Chlorophyll Remote Sensing Image for Fremont State Lakes

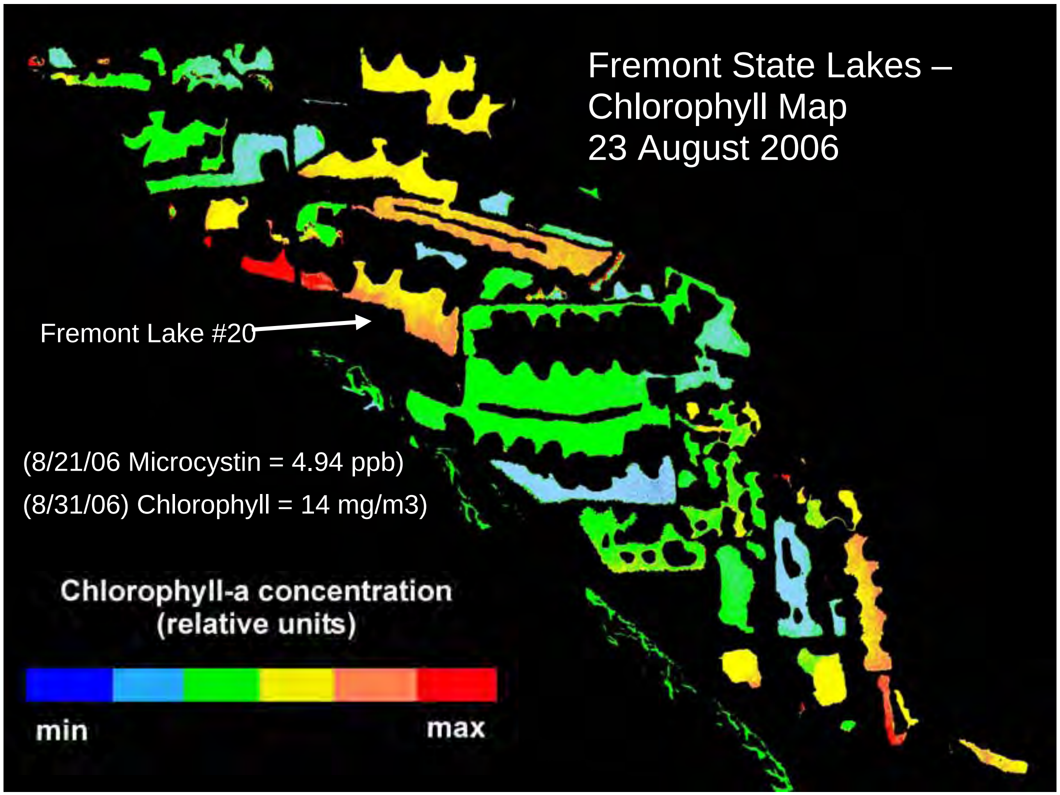


Figure 27. Phycocyanin Remote Sensing Image for Fremont State Lakes

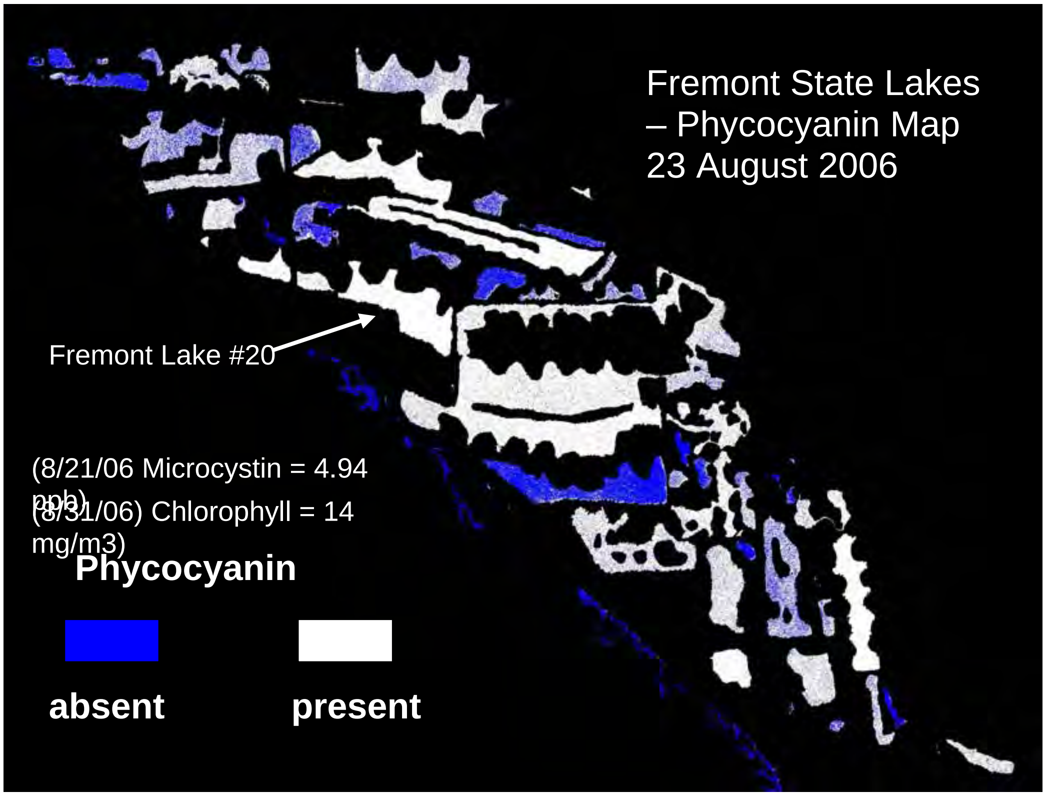
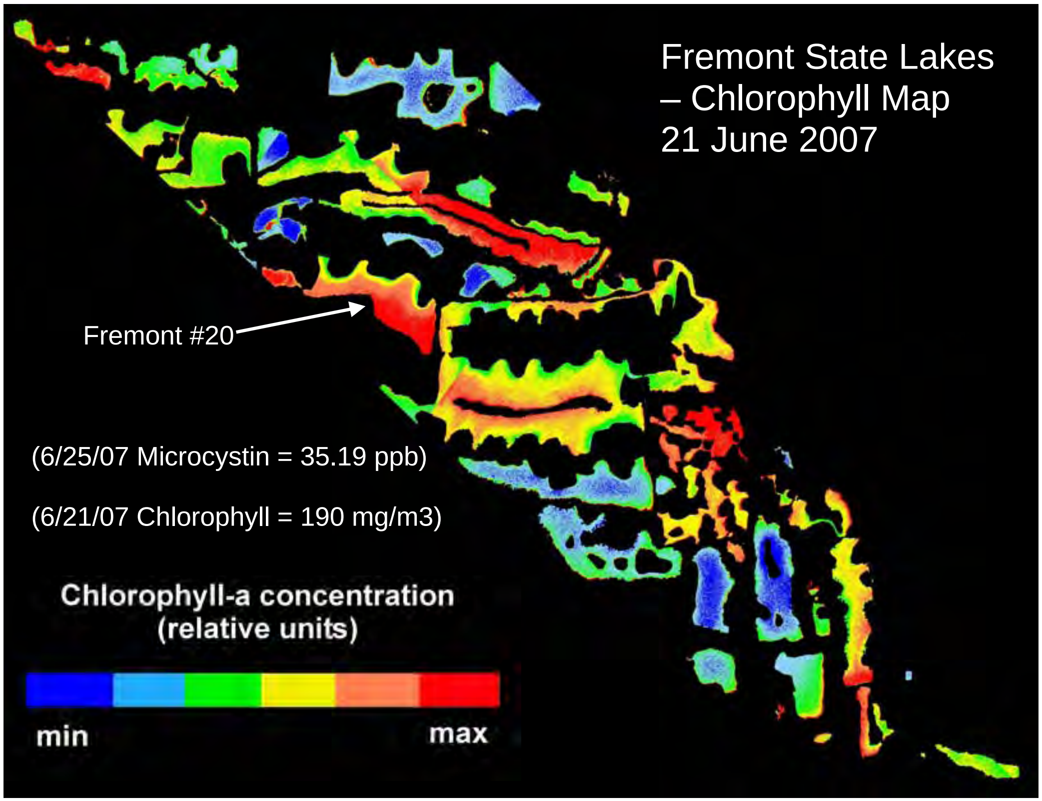


Figure 28. Chlorophyll Remote Sensing Image for Fremont State Lakes

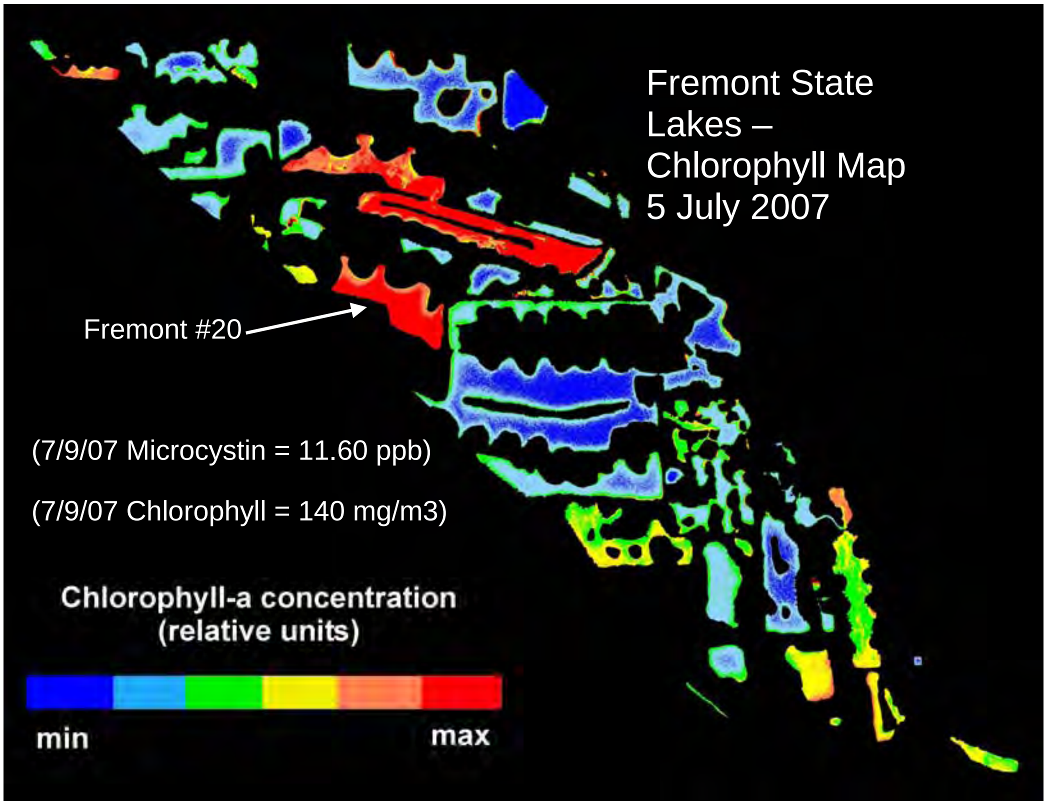


Fremont State Lakes – Chlorophyll Map 21 June 2007

(6/25/07 Microcystin = 35.19 ppb)

(6/21/07 Chlorophyll = 190 mg/m3)

Figure 29. Chlorophyll Remote Sensing Image for Fremont State Lakes



Fremont State Lakes – Chlorophyll Map 5 July 2007

(7/9/07 Microcystin = 11.60 ppb)

(7/9/07 Chlorophyll = 140 mg/m3)

Figure 30. Chlorophyll Remote Sensing Image for Fremont State Lakes

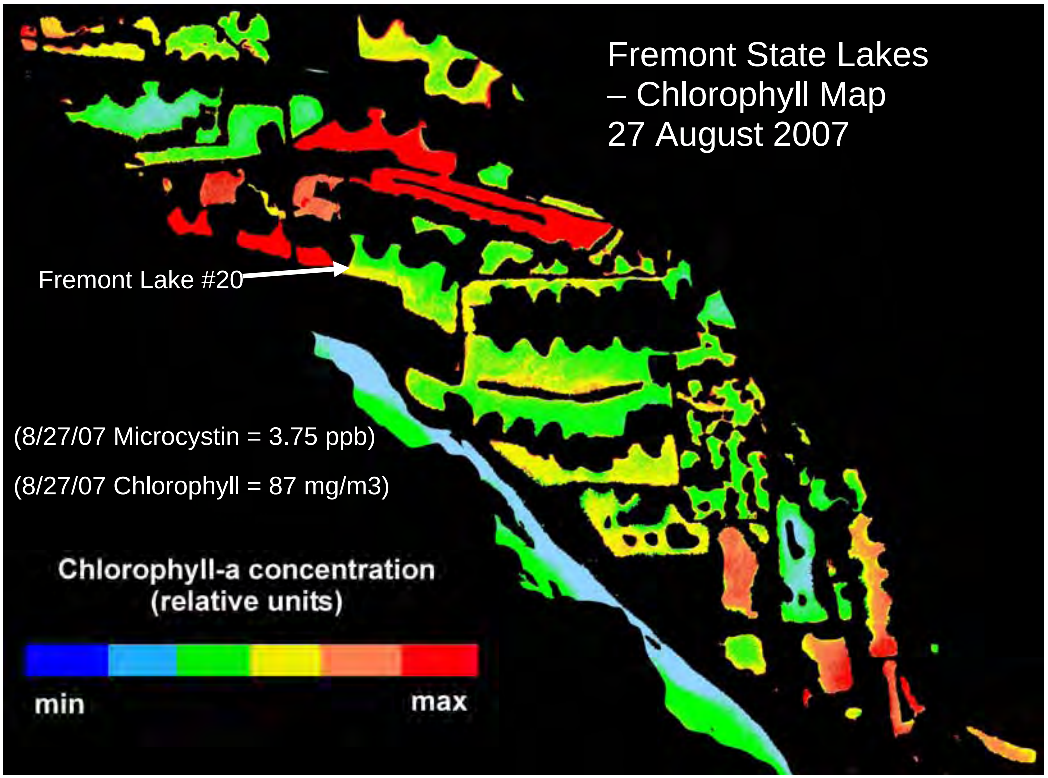


Figure 31. Chlorophyll Remote Sensing Image for Fremont State Lakes

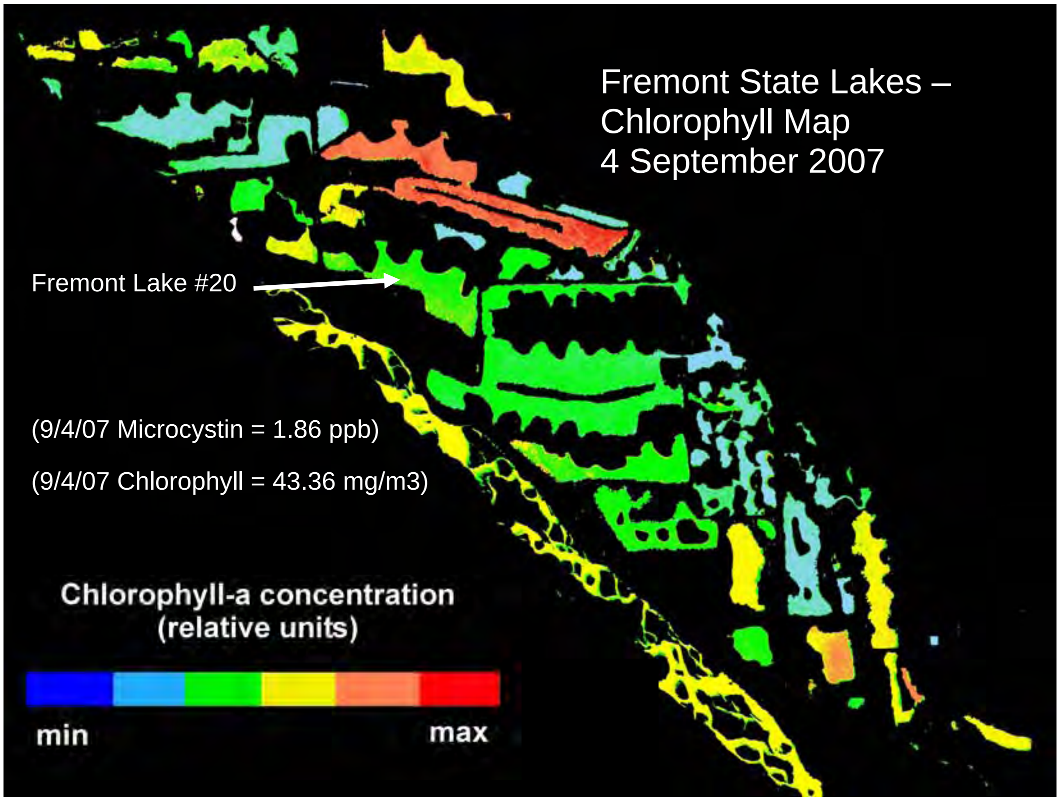


Figure 32. Phycocyanin Remote Sensing Image for Fremont State Lakes

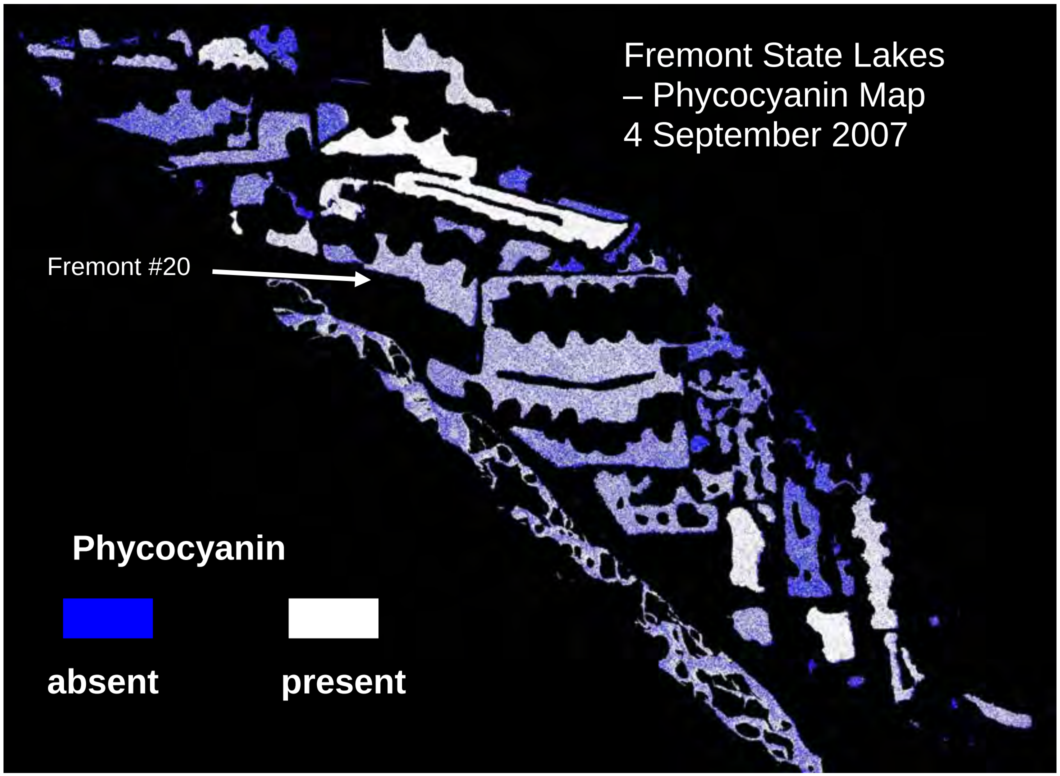


Figure 33. Chlorophyll Remote Sensing Image for Fremont State Lakes

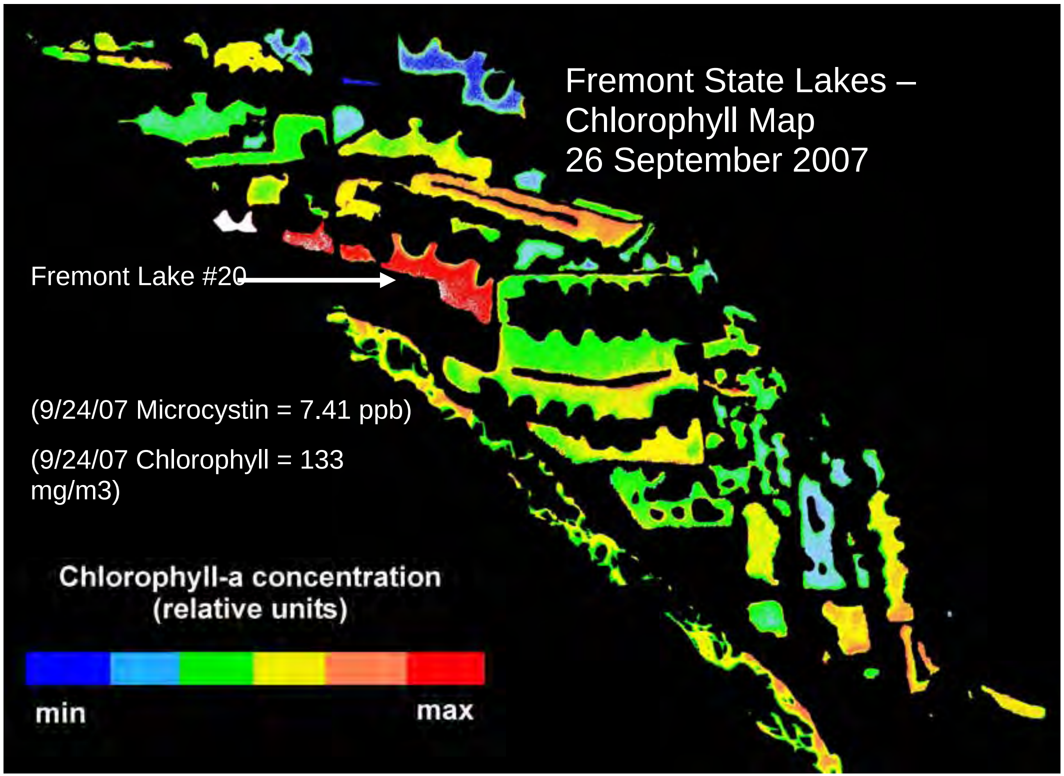


Figure 34. Phycocyanin Remote Sensing Image for Fremont State Lakes

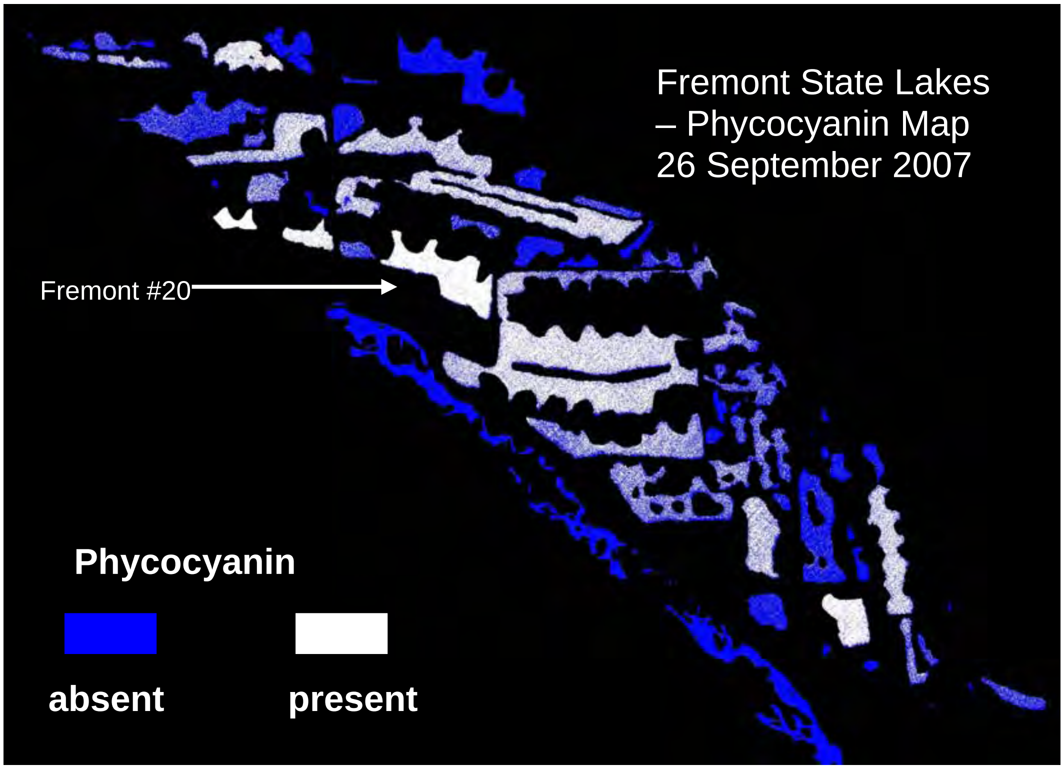


Figure 35. Chlorophyll Remote Sensing Image for Femont State Lakes

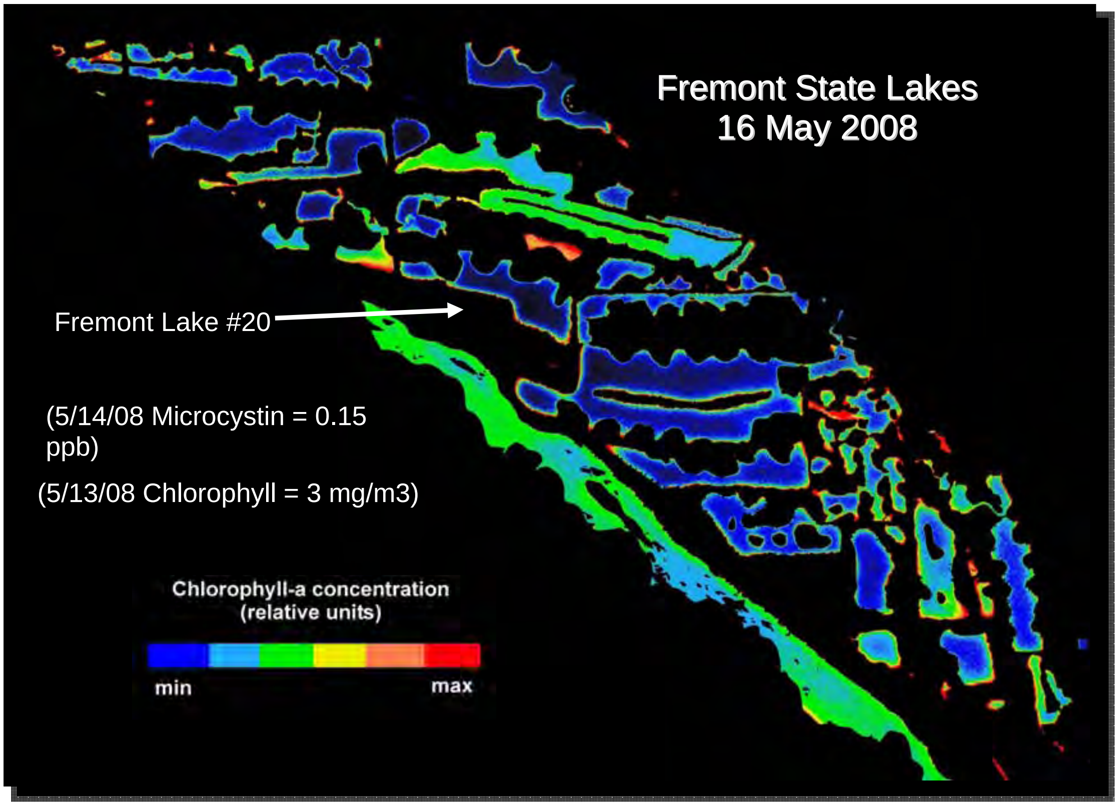


Figure 36. Pycocyanin Remote Sensing Image for Fremont State Lakes

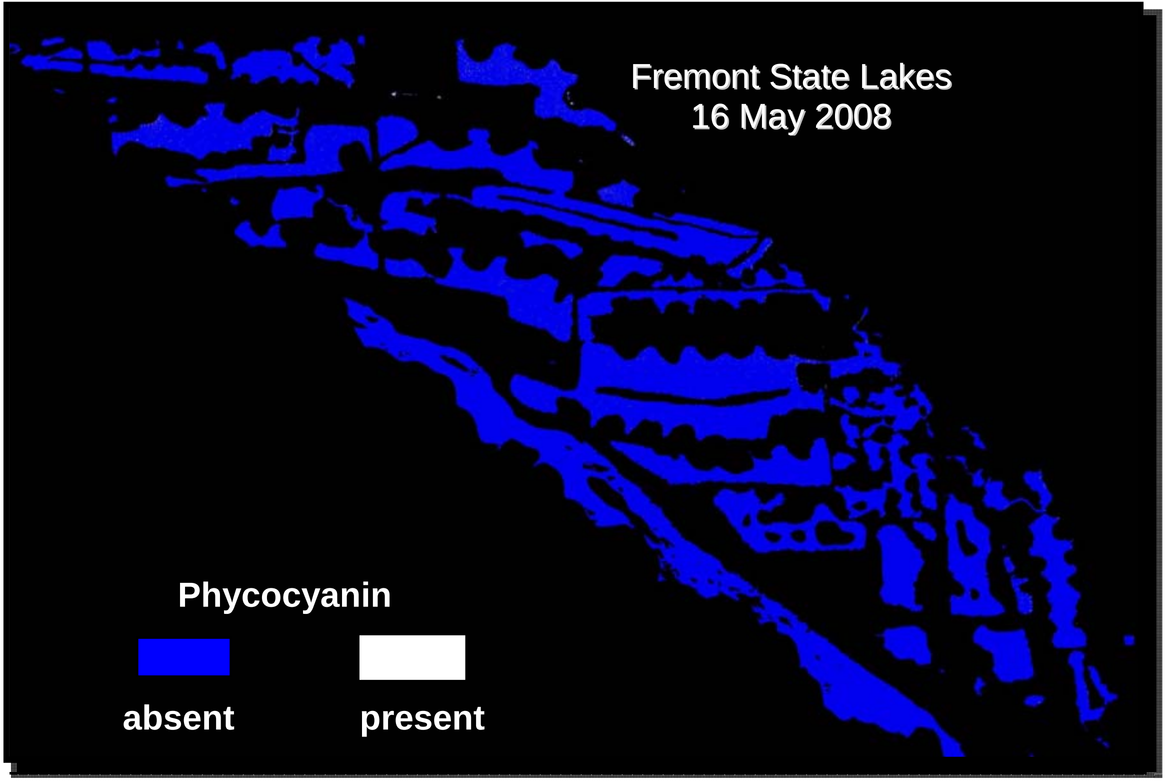


Figure 37. Chlorophyll Remote Sensing Image for Fremont State Lakes

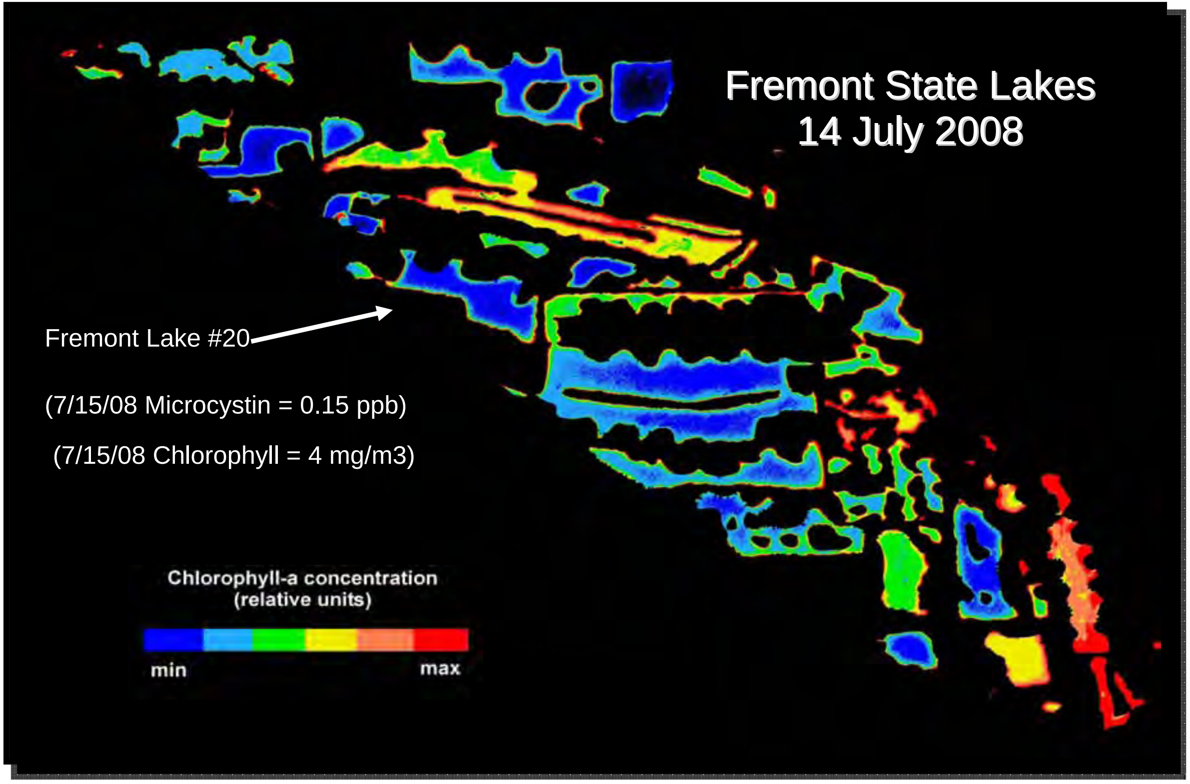


Figure 38. Phycocyanin Remote Sensing Image for Fremont State Lakes

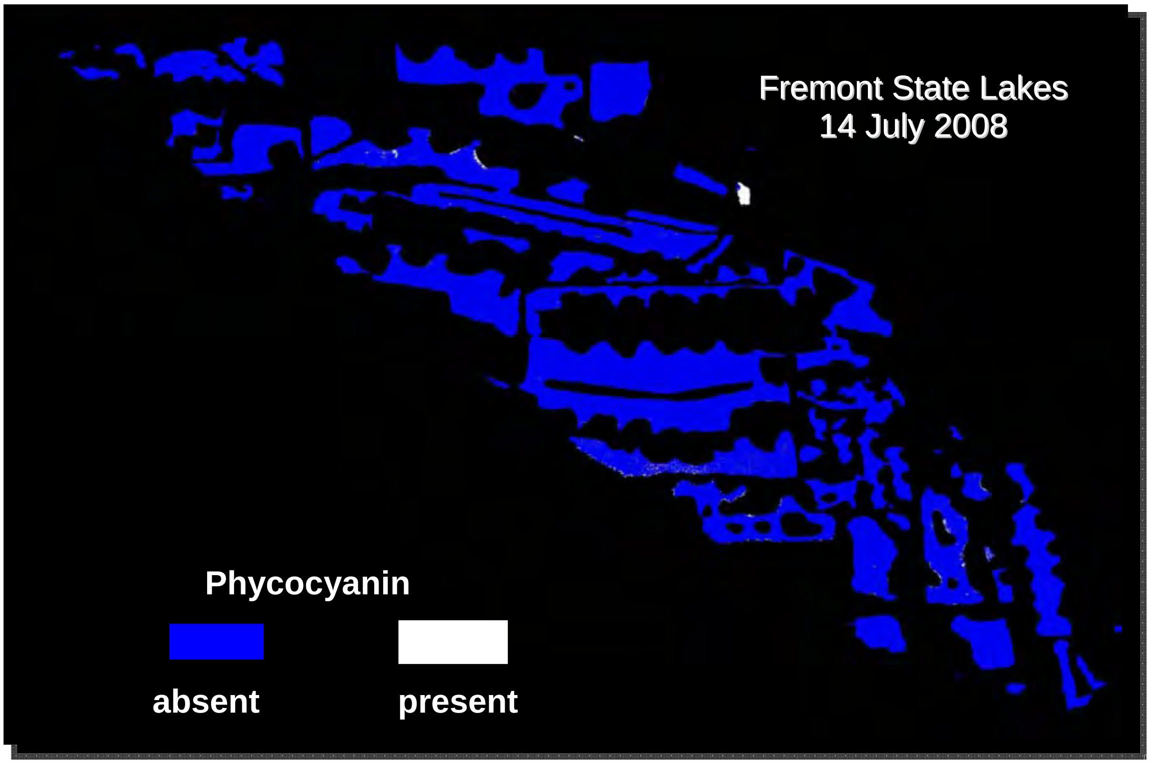


Figure 39. Chlorophyll Remote Sensing Image for Fremont State Lakes

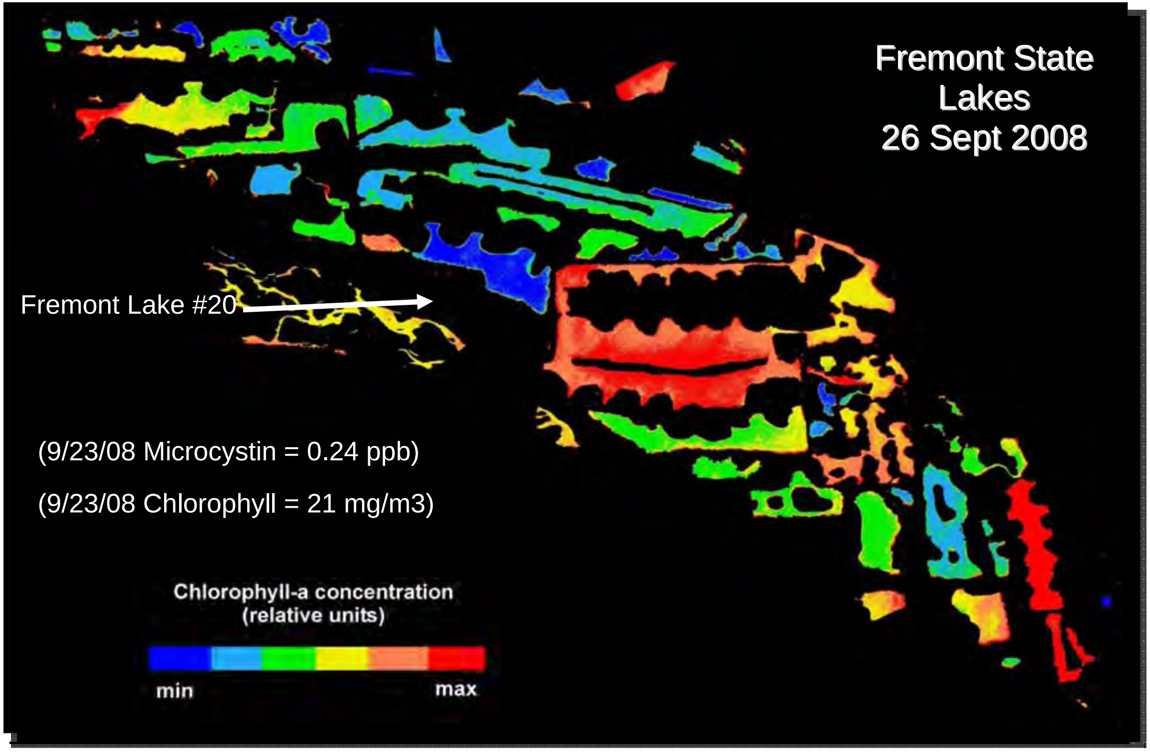
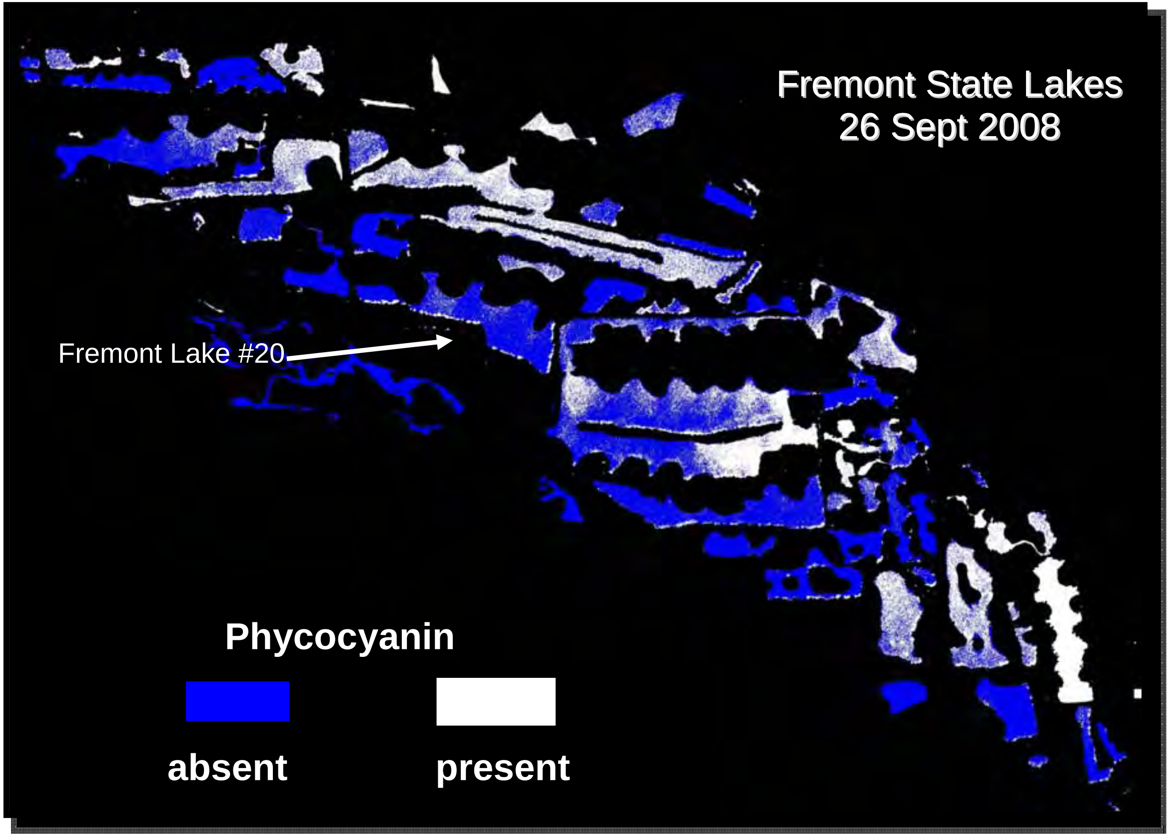


Figure 40. Phycocyanin Remote Sensing Image for Fremont State Lakes



Algae Bloom Characteristics

Chlorophyll data and remote sensing imagery was utilized to evaluate the extent, duration, magnitude, and frequency of algae blooms prior to and after the alum treatment (Table 5).

Bloom Extent

Remote sensing imagery suggests algal blooms at Fremont Lake #20 and surrounding lakes form quickly and typically impact the entire lake. Some spatial variation in chlorophyll was typically exhibited in the very early stages of a bloom. Sampling just off shore during these early bloom stages may provide data that is not representative of the entire lake.

Bloom Frequency and Duration

Chlorophyll *a* concentrations during 2005 indicate a bloom in late May that produced values greater than 100 mg/m^3. Values continued to increase to 170 mg/m^3 by August 30, 2005. This bloom encompassed approximately 101 days. Concentrations dropped to

117 mg/m^3 in early September only to increase to 168 mg/m^3 by the end of the sampling season on September 27, 2005. It is unknown how long the second bloom lasted.

Monthly chlorophyll sampling in 2006 was not sufficient to allow for an assessment of bloom duration. Conditions in 2007 were consistent with 2005 in that two primary blooms occurred. As in 2005, the first bloom occurred in late May producing chlorophyll *a* values up to 189.60 mg/m^3 by June 25. The spring bloom extended to approximately July 9, encompassing 49 days. The second bloom occurred in early August producing chlorophyll *a* concentrations as high as 88.88 mg/m^3 by August 6. The second bloom in 2007 lasted approximately 22 days.

Post alum treatment chlorophyll *a* concentrations measured in 2008 indicates two blooms, however, instead of a spring/summer bloom pattern that was noticed in previous years, both blooms in 2008 were during late summer. The first bloom took place around August 26, 2008 and lasted around eight days. This bloom produced chlorophyll *a* values as high as 20.50 mg/m3. The second bloom occurred on September 23 approximately 12 days after the end of the first bloom. The second bloom continued through the end of the monitoring season making it impossible to estimate bloom duration. In 2009, only one bloom was evident. A bloom started around August 5 and continued for approximately 28 days. The maximum chlorophyll concentration measured during this bloom was 30.74 mg/m^3.

Bloom Magnitude

Maximum pre-alum bloom concentrations of chlorophyll *a* ranged from 88.88 mg/m3 during the August 2007 bloom to 189.60 mg/m^3 during the May 2007 bloom (Table 5). The average chlorophyll *a* concentration for pre-alum blooms was 149.63 mg/m^3.

Maximum post alum bloom concentrations of chlorophyll *a* ranged from 21.40 mg/m^3 during the August 2008 bloom to 30.74 mg/m^3 during the August 2009 bloom. The average post alum treatment bloom concentration of chlorophyll *a* was 25.46 mg/m^3.

Table 5. Alage Bloom Characteristics at Fremont Lake #20

Bloom Start Date	Bloom Duration (days)	Bloom Maximum Chlorophyll *a* (mg/m3)	Bloom Extent
Pre-alum			
5/23/2005	101	170.40	Lake Wide
9/20/2005	Unknown	168.80	Lake Wide
5/22/2007	49	189.60	Lake Wide
8/6/2007	22	88.88	Lake Wide
Post Alum			
8/26/2008	8	21.40	Lake Wide
9/23/2008	7	24.24	Lake Wide
8/5/2009	28	30.74	Lake Wide

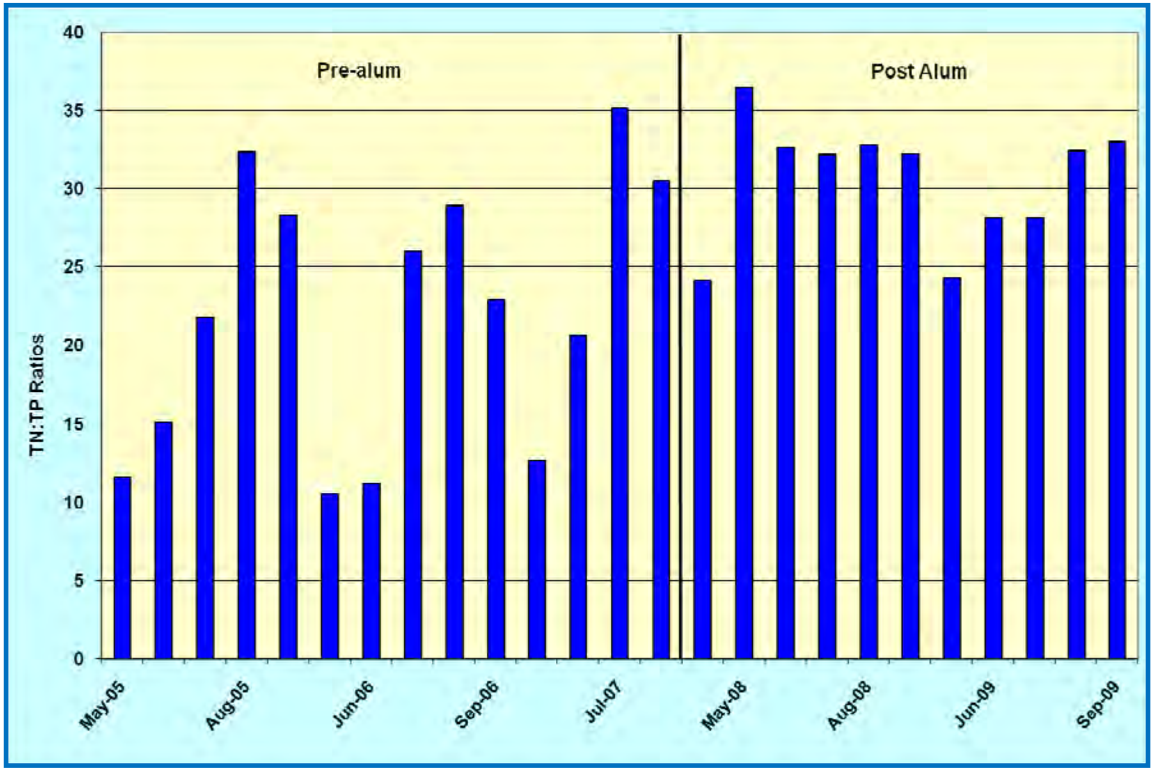
Total Nitrogen:Total Phosphorus Ratios

Total nitrogen to total phosphorus (TN:TP) ratios were calculated for all monitoring dates from 2005 through 2009 (Figure 41). Pre-alum TN:TP ratios ranged from 9.71 in May 2005 to 44.88 in August 2005. Post alum ratios ranged from 23.45 in May 2009 to 37.18 in May 2008.

The biggest shift in TN:TP ratios were for the month of May. Pre-alum TN:TP ratios in May 2005, May 2006, and May 2007 all dropped below 13, while the post alum ratios for May 2008 and May 2009 were greater than 24. The primary cause of increased post alum treatment TN:TP ratios were due to the large decrease in TP.

Literature suggests that generally lakes are phosphorus limited with TN:TP ratios >15 and nitrogen limited for TN:TP ratios <7. For ratios of TN:TP between 7 and 15, either P or N or both P and N could be limiting (1968, Vollenweider). Based on these guidelines, Fremont Lake #20 was phosphorus limited most of the time from 2005 through 2007, except for spring and early summer where phosphorus or nitrogen may have been the limiting nutrient. Ratios of TN:TP for all sampling dates in 2008 and 2009 suggest total phosphorus was the limiting nutrient. Low May TN:TP ratios during pre-alum sampling correspond to May blooms of blue green algae.

Figure 41. Monthly Average TN:TP Rations in Fremont Lake #20



Microcystin Predictors

The correlation between microcystin toxin concentrations and other factors such as TP, DOP, TN, chlorophyll *a*, and TN:TP ratios were evaluated using the Pearson Correlation. Microcystin concentrations showed little or no association with chlorophyll *a* (Pearson Correlation = 0.278), DOP (Pearson Correlation = 0.309), and TN (Pearson Correlation = 0.362). Microcystin concentrations did show a weak positive association with TP (Pearson Correlation = 0.591) and a weak negative association with TN:TP ratios (Pearson Correlation = -0.612).

Summary and Conclusions

The fish renovation, copper sulfate treatment, and alum treatment at Fremont Lake #20 has significantly changed water quality and the biological communities (Table 6). The goal of the treatments was to reduce microcystin toxin concentrations below beach posting criterion. This goal was achieved throughout 2008 and continued through 2010.

The alum was very effective in reducing in-lake TP concentrations but the large reduction in TN was not expected. These decreases may have been due, at least in part, to a decrease in nitrogen fixation based on a reduction in blue green biomass. Lower post treatment nutrient concentrations gave way to more algae taxa that included diatoms and green algae. While post treatment algae blooms were documented, bloom duration and magnitude were much less than for pre-treatment blooms. There was a decrease in oxygen demand near the lake bottom as near bottom oxygen concentrations were more than double what they were prior to the treatment.

While several parameters can be used to assess the potential for microcystin toxins, there is no single metric that can be used with confidence to predict toxin concentrations. In the absence of directly measuring toxin concentrations, algal community enumeration and identification, chlorophyll *a* densities, nutrient concentrations, TN:TP ratios, and phycocyanin presence can all be indicators of potential microcystin problems.

Since aluminum sulfate and sodium aluminate were both applied to the lake, pH was fairly stable during and after the application. The fish communities showed no signs of stress from the copper sulfate or alum treatments. Fremont Lake #20 will continue to be monitored to evaluate the longevity of the improvements.

As a result of the restoration activities and subsequent monitoring, Fremont Lake #20 was removed from Nebraska’s List of Impaired Waters in 2010. Fremont Lake #20 is now listed as a waterbody which fully supports all designated uses.

Table 6. Summary of Pre- and Post Alum Treatment Data

Parameter	**Pre-alum Sample Size**	**Pre-alum Average (2005-2007)**	**Post Alum Sample Size**	**Post Alum Average (2008-2009)**	**Percent Change**
TP Near Surface (µg/L)	46	128	38	22	- 83
TP Near Bottom (µg/L)	46	208	38	27	- 87
DOP Near Surface (µgL)	46	20	38	8	- 59
DOP Near Bottom (µg/L)	46	96	38	12	- 88
TN Near Surface (µg/L)	48	2601	43	838	- 68
TN Near Bottom (µg/L)	48	3111	43	870	- 72
Chlorophyll *a* (mg/m^3)	49	94.66	33	10.22	- 89
Water Transparency (inches)	49	16	32	99	+ 518
Microcystin Toxin (µg/L)	65	12.21	43	0.19	- 98
DO Near Surface (mg/L)	59	7.59	43	7.58	- < 1
DO Near Bottom (mg/L)	59	2.30	43	5.90	+ 256
pH Near Surface (*su*)	58	8.81	49	8.60	+ 2
pH Near Bottom (*su*)	58	8.12	49	8.52	+ 5
TN:TP Ratios (All May Dates)	11	12	7	33	+ 275
TN:TP Ratios (May-Sept)	49	23	32	32	+ 139
Algae Bloom Frequency (blooms/yr)	NA	2	NA	1-2	NA
Algae Bloom Magnitude Based on Chlorophyll *a* (mg/m3)	4	154.42	3	25.46	- 84
Algae Bloom Duration (days)	3	57	3	14	- 75

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