Final Technical Report

2013 Localized Treatment of Long Gulch Cove in Iron Gate Reservoir Using Environmentally Safe Algaecide

Prepared for

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1.0 INTRODUCTION

On February 18, 2010, the United States, the States of California and Oregon, PacifiCorp, regional Native American tribes, and a number of other stakeholder groups signed the Klamath Hydroelectric Settlement Agreement (KHSA). The KHSA lays out the process for additional studies, environmental review, and a determination by the Secretary of the Interior regarding whether removal of four dams owned by PacifiCorp on the Klamath River (i.e., J.C. Boyle, Copco 1, Copco 2, and Iron Gate dams) will advance restoration of the salmonid fisheries of the Klamath Basin and is in the public interest (which includes effects on local communities and tribes).

The KHSA includes provisions for interim operation of the dams and mitigation activities prior to potential removal of the hydroelectric facilities. One such provision—titled Interim Measure 11: Interim Water Quality Improvements—emphasizes water quality improvement projects in the Klamath Basin during the interim period.

Watercourse Engineering, Inc. (Watercourse), under contract to PacifiCorp, conducted a series of bench studies (in 2008, 2009, and 2011) to assess the potential use of algaecide as part of an overall algae management strategy in Copco and Iron Gate reservoirs on the Klamath River (Deas *et al*., 2009; Deas *et al*., 2012). These bench studies consisted of laboratory-based testing of two types of algaecide on collected samples of site-specific reservoir water to assess the effectiveness of algaecide application at different dosing conditions. The two tested algaecides included a copper-based algaecide, Algimycin PWF, and a hydrogen peroxide-based algaecide, GreenClean PRO. However, only the hydrogen peroxide-based algaecide is being assessed because it is deemed environmentally safe. Its active ingredient, hydrogen peroxide, is non-persistent and there is no bioaccumulation or sediment accumulation of the product because it degrades into water and oxygen, and this type of algaecide has been effective in controlling blue-green algae blooms and reducing microcystin concentrations (Matthijs *et al*. 2011). Further information on these tested algaecides is provided below in Section 2.2.

The bench studies indicated that algaecide could be effective in improving water quality by reducing algal concentrations and associated microcystin levels (microcystin is a toxin that can be produced by blue-green algae species). However, these lab-based bench tests were performed under controlled conditions that are not fully representative of in-situ conditions in the natural setting. As such, in September 2012, a limited pilot application of environmentally safe hydrogen peroxide-based algaecide was conducted in Copco Cove in Copco reservoir in order to evaluate the algaecide's effectiveness under the natural conditions of the reservoir (e.g., wind factors, advective influences, etc.). The 2012 pilot test application in Copco reservoir addressed several objectives:

- Define the necessary steps and activities associated with an in-situ algaecide application.
- Evaluate the effectiveness of algaecide in reducing algal cells by observing the response of chlorophyll *a* and cyanobacteria species to the algaecide application.

- Identify the effect of algaecide application on nutrient levels in the treated area of the reservoir.
- Determine the impact of algaecide application on microcystin concentrations.

Overall, the results of the 2012 pilot application study indicated that GreenClean Liquid is effective in reducing blue-green algae in the reservoir environment and reducing microcystin concentrations (Watercourse 2013). Response patterns of total nitrogen (TN) and total phosphorus (TP) concentrations mirrored the response of chlorophyll *a*, *Aphanizomenon flos-aquae*, and *Pseudoanabaena sp.* indicating that a large component of total nutrients were in their organic form at the time of treatment. Reductions in these constituents showed that the application of the hydrogen peroxide-based algaecide was effective in reducing the overall levels of algal cells. In addition, algaecide treatment led to modest increases in nitrate-plus-nitrite and phosphate concentrations. These increases were assumed to be a consequence of reduction in algal uptake and release of inorganic nutrients from algal cell death and lysis. However, ammonium levels were reduced. GreenClean Liquid application was also shown to reduce microcystin levels within the treated area of the reservoir. Observed algae reductions in 2012 were of short duration as algae were advected into the treatment area following treatment since the treatment area was not segregated from the remainder of the reservoir.

Based on the 2012 findings, as well as previous algaecide experiments, recommendations for future work included:

- Assessment of optimal algaecide application timing.
- Assessment of optimal algaecide application rates.
- Assessment of the effects of algaecide over time.
- Development of a plan that balances resources and appropriate level of monitoring inside the treatment area and in the non-treatment area during future algaecide applications.
- Evaluate the effectiveness of algaecide in reducing algal cells by observing the response of chlorophyll *a* and cyanobacteria species to the algaecide application.

In response to these recommendations, the 2013 study was implemented as described in this report. This study was completed in Long Gulch Cove in Iron Gate reservoir. A divider curtain was placed in the cove to isolate the treatment area (of approximately 7.5 acres) from the main reservoir area. All treatments in 2013 employed GreenClean Liquid, and associated monitoring was carried out.

This report is organized into several sections. Section 1 provides background information. Section 2 includes background information of conditions in the Klamath Basin, the use of algaecide treatment as a possible management strategy to reduce public health exposure, and previous algaecide studies. Section 3 describes methodology, including the study location, algaecide application procedures, and sampling procedures. Section 4 describes study results, followed by a discussion in Section 5. Section 6 summarizes conclusions and provides several recommendations for future consideration.

Section 7 includes references and there is an appendix that includes additional data in tabular and graphical form.

2.0 BACKGROUND

Detailed background information regarding algae production effects in the Klamath River, various algaecides, and the potential effects of algaecide are presented in Watercourse (2013). A brief summary is included herein.

2.1 Algae Production Effects in the Klamath River

Algae are a key component of aquatic systems, playing a vital role in food webs and producing oxygen through photosynthesis. However, excessive and/or persistent phytoplankton blooms can impair water quality. Algae can cause taste and odor problems in drinking water and can produce toxins that can affect wildlife, livestock, or humans via contact or ingestion. Algae can also present filter clogging challenges in water treatment and irrigation facilities and lower the aesthetic appeal and recreational use of surface waters. In addition, when toxins are involved, reservoirs and other surface waters may be posted with public health warnings, as has been the case with Copco and Iron Gate reservoirs and portions of the Klamath River.

The Klamath River is nutrient-enriched due to large loads of nutrients and organic matter to the river from hypereutrophic Upper Klamath Lake and other upstream sources. These nutrients help to cause seasonal algae blooms in the reservoirs along the Klamath River, including Iron Gate reservoir. Extensive seasonal algae standing crop have known direct effects on key water quality constituents in lakes and reservoirs, including dissolved oxygen (DO), pH, and nutrients, among others (Horne and Goldman 1994). The algal community in Iron Gate reservoir consists of diatoms, golden-brown algae, green algae, dinoflagellates, cryptomonads, microflagellates, and cyanobacteria (blue-green algae, BGA). Inter-annual variations are typical; as is the timing of the onset and decline of algae blooms (see Raymond 2008; 2009; 2010).

Cyanobacteria are of particular concern in reservoir management because they can produce undesirable toxins, including the hepatotoxin microcystin, which can, at a sufficient dose, affect the liver of animals, including humans. Cyanobacteria that can produce microcystin are *Microcystis aeruginosa*, *Anabaena flos-aquae*, *Planktothrix* (*Oscillatoria*), *Nostoc*, *Hapalosiphon*, *Anabaenopsis, and Pseudoanabaena* (World Health Organization (WHO) 1999; Oudra *et al*. 2002).

Characteristics of cyanobacteria that make their management challenging include the ability of these species to tolerate elevated water temperatures, reproduce at high rates, regulate their buoyancy, and, for certain species, the ability to fix atmospheric nitrogen. These characteristics can combine to create intensive bloom conditions for cyanobacteria populations. Heterogeneous (or “patchy”) distributions, accumulation of shoreline mats, wind driven accumulations, variability in toxin production, and other factors contribute to the management challenge.

2.2 Algaecides

Algaecides are a common technique for management and control of overabundant algae in ponds, lakes, and reservoirs (Cooke *et al*. 2005). Algaecides kill algae either by direct toxicity or through metabolic interference. Algaecide treatment can provide rapid removal of algae from the water column, sometimes resulting in dramatic short-term reductions in algal standing crop and improvements in water clarity. Application frequency is a function of the lake or reservoir management objective (e.g., nuisance, taste and odor issues, toxin management, recreation, etc.) and the type of algaecide used. In certain cases, algaecides are applied annually but are, more typically, applied several times throughout periods of algae growth to prevent or reduce algal blooms.

There are various types of algaecides available commercially. Algaecides that have been identified as being potentially useful for water quality improvements in the Klamath River fall into two major categories: copper-based and peroxide-based. For this study, a peroxide-based algaecide was applied.

Various studies have shown that peroxide-based algaecide is potentially a safer and equally-effective alternative to copper (Drábková *et al*. 2007; Barrington and Ghadouani 2008; Fan *et al.* 2014). Hydrogen peroxide is non-persistent and there is no bioaccumulation or sediment accumulation of the product because it degrades into water and oxygen (Ding *et al*. 2012; Environmental Protection Agency (EPA) 2012). Furthermore, hydrogen peroxide exists naturally in lacustrine environments where it is generated photochemically from organic products in the presence of sunlight (Cooper and Zika 1983; Scully *et al*. 1995; 1996). Given these attributes, hydrogen peroxide is environmentally benign (Antoniou *et al*. 2005; Qian *et al*. 2012).

The mechanism by which hydrogen peroxide breaks down cyanobacteria has been the subject of several studies. When applied, hydrogen peroxide acts as an oxidizing agent that inhibits algal growth by altering algal physiological and biochemical processes (Samuilov *et al*. 2004; Qian *et al*. 2010; 2012). Additionally, Ross *et al*. (2006) established that hydrogen peroxide addition elicited caspase activity (e.g., programmed cell death) in *Microcystis aeruginosa*. More recently, Ding *et al*. (2012) observed that hydrogen peroxide induces apoptotic-like programmatic cell death[1] (PCD) in *Microcystis aeruginosa* (see also Ross *et al*. (2006)).

Recent studies have observed changes in physiological parameters of algae associated with the introduction of hydrogen peroxide. These include changes in algal mortality, chlorophyll content, cellular soluble protein, microcystin synthesis, and photosynthetic activity (Drábková *et al*. 2007; Ding *et al*. 2012; Qian *et al*. 2012. In addition, Qian *et al*. (2010) demonstrated that hydrogen peroxide inhibited carbon assimilation thereby inhibiting algal growth. Furthermore, hydrogen peroxide also decreases the levels of photosynthetic pigments: chlorophyll *a*, phycocyanobilin, allophycocyanin, and phycoerythrin (Qian *et al*. 2010). These pigments capture light energy necessary for

[1] Apoptotic-like PCD is a pattern of cell death affecting single cells, marked by shrinkage and fragmentation of the cell into membrane-bound bodies that are eliminated by phagocytosis (ingestion by other cells, such as microphages).

photosynthesis, and so reduction of their levels inhibits algal growth. Another way that hydrogen peroxide inhibits growth is by changing the rhythms of cyanobacterial timing genes. Many physiological and metabolic activities that occur, such as cell division, nitrogen fixation, photosynthesis, carbon uptake and the biosynthesis of secondary metabolites, are controlled by these rhythms. Hydrogen peroxide has been observed to affect circadian rhythms in cyanobacteria. Some observed impacts are declines in solar energy utilization and the synthesis of carbohydrates and high energy molecules, which are necessary for cyanobacterial growth (Qian *et al*. 2012). Qian *et al*. (2010) also showed that hydrogen peroxide reduces or inhibits the production or synthesis of microcystin. Finally, hydrogen peroxide can also destroy toxins that are released upon the lysis of cyanobacterial cells (Svrcek and Smith 2004).

In the 2008 bench study, two hydrogen peroxide-based algaecides, GreenClean PRO and PAK-27, were tested in a bench-top setting with water samples taken from Copco reservoir. In 2009, further bench tests were performed to investigate the effects of higher dosages and re-application of GreenClean PRO on the algae species present in Copco reservoir. The performance of the liquid version of GreenClean PRO, GreenClean Liquid, was tested in 2011. Findings from these studies can be found in Deas *et al*. (2012). Based on the results of these previous studies, GreenClean Liquid was chosen for the 2012 Copco Cove (Watercourse 2013) and 2013 Long Gulch Cove (Iron Gate reservoir) in-reservoir studies.

GreenClean Liquid, like GreenClean PRO, is produced by BioSafe Systems, LLC, and is a hydrogen peroxide-based alternative to copper-based algaecide and algaecides with other toxic chemicals as their active ingredient. In California and Oregon, there are no known runoff or usage restrictions associated with the use of GreenClean Liquid, which utilizes sodium carbonate peroxyhydrate (SCP) as its active ingredient. SCP is a stabilized form of hydrogen peroxide that is paired with peroxyacetic acid (PAA). PAA is a compound made up of hydrogen peroxide and acetic acid. This compound is an activated form of hydrogen peroxide and acts as a more stable and powerful oxidizer (Larose *et al*. 2008). The combination of hydrogen peroxide and PAA causes an oxidation reaction that breaks down or damages algae cell walls (Knox 2009). The reaction works quickly (seconds to minutes), reducing the likelihood of mutational resistance. As the reaction takes place, hydrogen peroxide and PAA break down into natural compounds: water, oxygen and elements of organic acids (Larose *et al*. 2008; EPA 2012). Like hydrogen peroxide, PAA does not persist in the environment (Knox 2009). Further, the concentration of PAA in GreenClean Liquid is extremely low (on the order of 0.0000033-0.000083 molar for the manufacturer's listed range of application rates) (V. Choppakatla, pers. comm.).

2.3 Consideration of Potential Algaecide Effects

Potential algaecide effects include impacts on other plants and fish, dissolved oxygen, nutrient concentrations, and toxins. Use of algaecides can cause temporary effects on non-target plants, but recovery of those plant communities is usually rapid (Wagner 2004). The use of algaecides above their recommended dosages may impact fish species. However, the EPA fact sheet states that when SCP "is applied in accordance with

directions on the label, no harm is expected to birds, other terrestrial animals, freshwater fish, or freshwater invertebrates" (EPA 2002), and several algaecides are designed for use in the treatment of fish ponds and other water bodies that contain fish (BioSafe 2012). Oxygen depletion in the water column may follow algaecide application due to the decomposition of dead algae. Nutrient concentrations can increase or decrease following algaecide applications. Also, because cyanotoxins are stored intracellular, algaecide treatments could lead to releases of intercellular toxin to surrounding waters (Kenefick *et al*. 1993; Jones and Orr 1994; Touchette *et al*. 2005).

More recent studies specific to the application of H2O2 examine toxin release associated with cell lysing and the fate of microcystin. Fan et al. (2013) presents information on cell lysing and damage, identifying that H2O2 application damages only a portion of the cells and many remain intact. Recent research (Barrington, et al. 2013, Matthijis, et al. 2011) has also indicated that hydrogen peroxide application to cyanobacteria blooms can rapidly reduce both cyanobacteria (as indicated by chlorophyll *a*) and microcystin concentrations in water bodies while promoting more favorable phytoplankton assemblages. These studies are consistent with the idea that hydrogen peroxide, a strong oxidant, is able to oxidize microcystin during or immediately following cell lysis. Barrington et al. (2013) reported that while cell lysing occurred with H2O2 application, total microcystin was reduced for up to three weeks following treatment. Further, dissolved microcystin continued to decrease to non-detectable levels a few days after treatment. Because H2O2 oxidizes out the system quickly (e.g., hours), these declines in microcystin concentrations may be due to UV radiation, bacterial activity or other environmental factors.

Hydrogen peroxide-based algaecides can reduce dissolved microcystin through several mechanisms. Oxidation due to hydrogen peroxide treatment can directly reduce dissolved microcystin, and Qiao *et al*. (2005) identified that reductions are markedly increased where ultraviolet light (UV) is present. Matthijs et al. (2011) states that H2O2 has a strong oxidizing ability to help break down microcystin, but is also assisted by light (UV). Other studies indicated that H2O2 can help speed up the degradation of microcystin in the presence of ultraviolet light (Bandala et al. 2004; Cornish et al. 2000). The persistence of hydrogen peroxide in aquatic environments is short, which may limit the effectiveness of H2O2 at degrading microcystin from recently lysed cells (Fan *et al*. 2013, Qiao *et al.* 2005). Other research (Lawton et al., 1999; Liu et al., 2002; Rodrıguez et al., 2007, 2008) has demonstrated that the products of the oxidation of forms of microcystin (-LR and –RR congeners) are non-toxic, and thus no longer present a danger for public health. Though H2O2 treatment may lead to cell lysing, this in itself does not increase total microcystin.

Other concerns expressed with algaecide treatment has been that nutrients will be released as a result of cell lysis and contribute to additional algal growth. Nutrient release upon cell lysis and cell death will occur with any algaecide or pesticide application. Additionally, algal growth in Klamath River reservoirs is not nutrient-limited such that additional bioavailable nutrients would exacerbate seasonal algal conditions. Further, cell lysis will result in some algal biomass sinking to the reservoir bottom, where nutrients within the biomass will not be available in the photic zone for uptake as algal biomass.

Regardless, in the worst case, released nutrients would only be able to form the same amount of cyanobacteria that was eliminated through treatment.

As discussed in the Introduction, the algaecide studies conducted to date (Watercourse (2013) and as described herein) have included objectives aimed at assessing both the benefits and other potential water quality impacts of algaecide use. As such, in addition to assessing the effectiveness of potential algaecide treatment in reducing algal standing crop, the studies have also investigated the effects of algaecide application (such as described above) on nutrient and microcystin concentrations in the surrounding water. Study results pertaining to these various objectives are described further in Sections 4 and 5 of this report.

3.0 METHODOLOGY

This section describes the study location, algaecide application procedure, and the sampling methods associated with the in-situ pilot application of GreenClean Liquid, which utilizes SCP as its active ingredient, conducted in Long Gulch Cove of Iron Gate reservoir in September and October of 2013.

The 2013 pilot study in Long Gulch Cove utilized the application and monitoring plan developed for the 2012 pilot study, with slight adjustments (Watercourse 2013). The same methodologies from 2012 were used in 2013, with a few modifications and additions (detailed below).

3.1 Study Location

Long Gulch Cove (Figure 1) in Iron Gate reservoir was selected as the study location based on its size, accessibility, and the amount of algae observed. Conducting the study in Long Gulch Cove utilized the natural shape of the cove to limit water movement and potential exposure to wind. Additionally, a divider curtain was deployed in the cove to isolate the treatment area (of approximately 7.5 acres) from the main body of the reservoir. The curtain, made of Type 2 DOT, yellow vinyl-coated polyester, was assembled in place in sixteen 50 foot sections with a total length of 800 feet (Figure 2). Each of the sixteen sections was fabricated to extend from surface to a maximum depth of approximately 35 feet. The curtain was deployed using surface floats and anchors to maintain position.

Figure 1. Aerial photo of Iron Gate reservoir that includes the location of Long Gulch Cove.
Courtesy of Google Earth

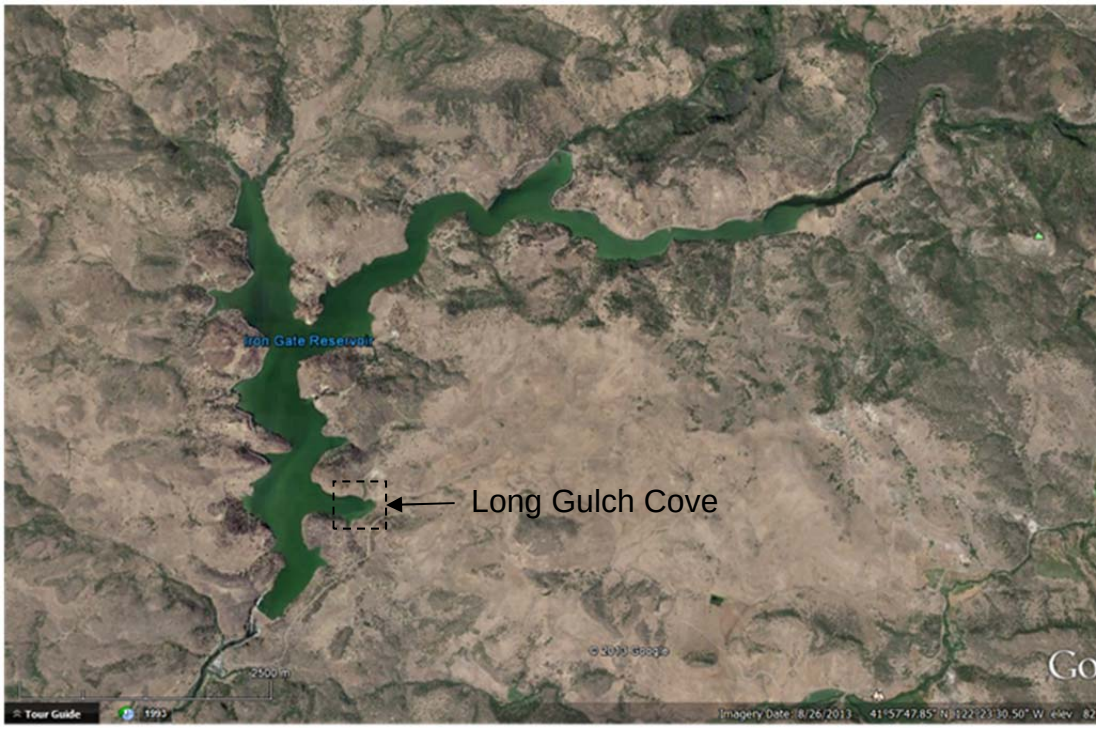


Figure 2. Curtain installed to isolate a portion of Long Gulch Cove (looking from the southern anchor point towards the northern anchor point).

3.2 Algaecide Application Procedures

The algaecide used for the study was GreenClean Liquid (EPA Registration No. 70299-2), which is manufactured by BioSafe Systems, LLC (BioSafe). The algaecide application was performed by Clean Lakes, Inc. (CLI) on September 11, 2013 and October 2, 2013. The application of algaecide was conducted in compliance with:

- California Department of Pesticide Regulation (DPR).
- State Water Resources Control Board (SWRCB) Water Quality Order No. 2004-0009-DWQ, which is the Statewide General National Pollutant Discharge Elimination System Permit for the Discharge of Aquatic Pesticides for Aquatic Weed Control in Waters of the United States (General Permit No. CAG990005).

GreenClean Liquid, contained in either 275-gallon totes (September 11, 2013) or 55-gallon drums (October 2, 2013), was delivered to the Pacific Power facility in Yreka, California on the mornings of September 11, 2013 and October 2, 2013. CLI staff delivered the totes/drums to the Long Gulch Cove boat ramp. At the boat ramp, CLI staff transferred GreenClean Liquid from the delivery totes/drums to the treatment vessel using a closed system algaecide transfer procedure.

On the morning of September 11, 2013 and October 2, 2013, the information board located at Long Gulch Cove was posted with a notice informing the public that algaecide application was taking place. These postings were removed the following morning after post-event monitoring was completed.

Algaecide application at Long Gulch Cove occurred from 11:00 am to 12:50 pm on September 11, 2013 and from 11:20 am to 12:20 pm on October 2, 2013 (Table 1). CLI utilized a LittLine® Littoral Zone Treatment vessel for algaecide application. The isolated area of Long Gulch Cove covered approximately 7.5 acres of water surface area. On September 11, 2013 the upper 6 feet (1.8 meters) of the water column were treated, leading to a treatment volume of 45 acre-feet. A total of 405 gallons of GreenClean Liquid was applied, which amounts to 9.0 gallons per acre and an active ingredient concentration of 7.50 ppm. This dosage corresponds to the specimen label application rate for medium density (filamentous) algae conditions (BioSafe 2012). Also on September 11, 2013, an additional 7 feet (2.1 meters) of the water column was treated (from 6 feet to 13 feet below the surface), leading to a treatment volume of 48.3 acre-feet. A total of 145 gallons of GreenClean Liquid was applied, which amounts to 3.0 gallons per acre and an active ingredient concentration of 2.50 ppm. This dosage corresponds to the specimen label application rate for low density (filamentous) algae conditions (BioSafe 2012). Therefore, on September 11, 2013, a two-step application was conducted; a higher algaecide concentration was applied to the upper 6 feet of the water column and a lower concentration was applied at a depth of 6 to 13 feet below the surface.

On October 2, 2013 GreenClean Liquid was applied to the upper 4 feet (1.2 meters) of the water column were treated, leading to a treatment volume of 30 acre-feet. A total of 90 gallons of GreenClean Liquid was applied, which amounts to 3.0 gallons per acre and

an active ingredient concentration of 2.50 ppm. This dosage corresponds to the specimen label application rate for low density (filamentous) algae conditions (BioSafe 2012).

After algaecide application, empty algaecide drums were triple rinsed into the application vessel's pesticide tanks. Rinsed drums were then transported by CLI to their disposal facility in Martinez, CA for removal per DPR regulations.

Table 1. Treatment dates, depths, areas, volumes, and concentrations.

Date	Treatment Depth (ft/m)	Treatment Area (acre-ft)	Treatment Volume (gal)	Treatment Concentration (ppm)
09/11/2013	0-6 ft (0-1.8 m)	45.0	405	7.50
09/11/2013	6-13 ft (1.8-4.0m)	48.3	145	2.50
10/02/2013	0-4 ft (0-1.2 m)	30.0	90	2.50

3.3 Sampling Methods

Grab samples and physical measurements were collected at six locations, which include three non-treatment sites (identified with a "N") located outside the treatment area to represent untreated conditions, and three treatment sites (identified with a "T") located within the treated area (Figure 3). Non-treatment sites were located approximately 250 feet from the curtain isolating part of Long Gulch Cove (actual distances varied slightly). The non-treatment sites were able to freely mix with water from the rest of Iron Gate reservoir. Hydrogen peroxide, the active ingredient in the algaecide, was collected at three treatment locations (T1, T2, and T3) inside the curtain. At each location, samples were collected at two depths: near surface (0.1 m depth) and an integrated sample (surface to approximately 0.5 m above bed or 6 m, whichever was less).

The sampling locations were identified using a Garmin Oregon® 450 Geographic Positioning System (GPS) prior to pre-treatment sampling. The coordinates were recorded in the GPS and later used to position the boat when samples were taken. A summary of the sampling location coordinates are included in the attached Appendix A. This procedure ensured consistent repositioning at each sampling location on the reservoir where pre-event (representing background conditions), immediately after event/treatment (event), the next day (post-event), and one-week after treatment (one-week) samples were collected.

Figure 3. Approximate sampling locations for in-situ algaecide study in 2013. "N" and "T" were used to denote non-treatment and treatment sites, respectively. "N Curtain" denotes the approximate northern anchor point for the curtain, while "S Curtain" denotes the southern anchor point. The dashed line represents the approximate location of the curtain in Long Gulch Cove.

Samples were collected from a boat using a Cole-Parmer Masterflex® E/S portable sampler, which is a variable speed peristaltic pump used in conjunction with a ¼ inch tube to draw water from approximately 0.1 m into a 14-liter churn splitter. At each location prior to sample collection, the hose was rinsed with environmental water by running the pump for 1 minute before collecting samples. The churn splitter was also triple rinsed with environmental water based on standard procedures. Following the environmental rinses, the churn splitter was filled with sample water and the prepared sample bottles were filled from the churn splitter as per standard operating procedures.

An integrated tube sampler was used to collect water samples from the top 6 m (or less if the water column was less than 6.5 m) of the water column. A 1.5-inch inside diameter, silicon tube was lowered vertically through the water column with both ends open. When the target depth was reached, the above water end of the hose was clamped shut and the submerged end was slowly raised to the water surface. The content of the integrated tube sampler was then discharged into a 14-liter churn splitter located on the boat.

At each sampling location and depth, grab samples were collected for subsequent laboratory analysis of nutrients, microcystin, cyanobacteria (blue-green algae) species enumeration, and chlorophyll *a*. Hydrogen peroxide samples were also collected at treatment locations (T1, T2, and T3). In addition to these grab samples, measurements of

water temperature, DO, pH, electrical conductance, turbidity, Secchi depth, and total depth were taken at each location.

Laboratory analysis of samples were performed for total Nitrogen (TN), nitrate and nitrite (NO3 + NO2), nitrite (NO2), ammonia (NH4), total Phosphorus (TP), phosphate (PO4), dissolved organic carbon (DOC), chlorophyll *a,* microcystin, and hydrogen peroxide. Samples were delivered directly to the Biogeochemistry Laboratory (P.I. Dr. Randy Dahlgren) at University of California, Davis within 48 hours. Samples analyzed for microcystin were frozen and shipped overnight to Tamarack Environmental Laboratories, LLC, Washington, Michigan. Hydrogen peroxide samples were transported to Davis, California and then by courier to the McCampbell Analytical, Inc. laboratory in Pittsburg, California within the 7-day holding time. All samples were stored and transported or shipped on ice. Laboratory information associated with each constituent is included in Table 2.

Table 2. Laboratory methods, method detection limits (MDL), and reporting limits (RL), as applicable for each water quality constituent.

Constituent	Units	Method	Preservative	MDL[a]	RL[a]	Laboratory
TN	mg/l	NEMI[b] I-4650-03	None	0.01	0.02	Biogeochemistry Laboratory, U.C. Davis
NO3+NO2	mg/l	Nitrate via V(III) reduction[c]	None	0.005	0.01	Biogeochemistry Laboratory, U.C. Davis
NO2	mg/l	Nitrate via V(III) reduction	None	0.002	0.01	Biogeochemistry Laboratory, U.C. Davis
NH4	mg/l	SM[d] 4500-NH3 F	None	0.005	0.01	Biogeochemistry Laboratory, U.C. Davis
TP	mg/l	NEMI I-4650-03	None	0.01	0.01	Biogeochemistry Laboratory, U.C. Davis
PO4	mg/l	SM 4500-P E	None	0.001	0.005	Biogeochemistry Laboratory, U.C. Davis
DOC	mg/l	EPA 415.3	None	0.1	0.1	Biogeochemistry Laboratory, U.C. Davis
Chlorophyll *a*	µg/l	EPA 445.0	None	1ppb	1 ppb	Biogeochemistry Laboratory, U.C. Davis
Microcystin	mg/l	ELISA[f]	None	0.16	n/a	Tamarack Environmental Laboratory
Hydrogen Peroxide	mg/l	Titanium Sulfate/ Spectrophotometric[g]	None	n/a	1.0	McCampbell Analytical, Inc.

[a] Units are in mg/l unless otherwise specified.

[b] National Environmental Methods Index.

[c] This method was developed by UC Davis Department of Land, Air and Water Resources (Doane and Horwath 2003).

[d] Standard Methods.

[e] Environmental Protection Agency.

[f] USEPA Region 9 SOP 1305 (Envirologix ELISA method).

[g] This method is from Industrial and Engineering Chemistry Analytical publication, "Colorimetric Determination of Hydrogen Peroxide" (Eisenberg 1943).

4.0 RESULTS

The average water quality sample results (including microcystin, hydrogen peroxide, and nutrients), turbidity and Secchi disk measurements, and algal response from the September 2013 and October 2013 test applications of an environmentally safe algaecide in Long Gulch Cove are summarized below and the discussion of these results is presented in Section 5. Individual sample site measurements for water temperature, DO, pH, turbidity, Secchi depth, and total water depth are included in Appendix A. While results are presented in combination herein, the September and October treatments are considered separate activities for the purpose of this study. As noted below, algal,

meteorological, water temperature, and other conditions were notably different during the September and October period that make direct comparison unrealistic.

4.1 Visual Observations

Visual observations in and around the study area identified high algal densities, principally *Microcystis aeruginosa*, throughout the study period. Weather conditions were variable between treatment events and sampling periods. During the September 2013 application event, weather conditions were calm and clear with extensive algae presence (Figure 4). One week later, weather conditions were still clear, but windy. During the October 2013 event and subsequent sampling, weather conditions were colder, clear with some clouds, and windy.

As previously discussed, a divider curtain was installed to isolate Long Gulch Cove from the rest of Iron Gate reservoir. Visual observation indicated that the curtain was effective at isolating the cove water. In September, the surface area behind the curtain (within the cove) had a noticeably different appearance than the adjacent surface area (Figure 5). This was consistent throughout the September and October sampling events. The distribution of algae varied throughout the study period, ranging from uniformly high densities to heterogeneous or patchy conditions. A general observation was that subsequent to treatment, clarity was notably improved. While algae were still present, accumulations of algae in surface waters were dispersed and small clumps of what appeared to be dead algae were present.

Figure 4. Surface algae conditions in the study site prior to algaecide application (September).

Figure 5. Long Gulch Cove treatment area prior to treatment. September 11 (left) and September 18 (right).

4.2 Water Quality

Water quality consisted of vertical profiles measured with a water quality probe, and grab samples. Samples were collected at all non-treatment and treatment locations pre-event, during event (shortly after treatment), post-event (one day), and approximately one week after treatment. Pre-event, post-event, and one-week sampling was carried out at approximately the same time on each day to provide for a direct comparison (however, some sampling occurred at different times) (Table 3). Vertical profile measurements were completed at 0.5 meter increments. Two grab samples were collected from each site; one was from just below the surface of the water (approximately 0.1 m) and the other was an integrated sample. The integrated sample contained water from the surface to 0.5 m above the bed or 6.0 m (whichever was less). All field data are included in Appendix A.

Table 3. Approximate sampling time range for September and October, treatment and non-treatment sites.

	September Samplings			October Samplings		
Sample	**Date**	**Treatment**	**Non-Treatment**	**Date**	**Treatment**	**Non-Treatment**
Pre-Event	09/11/2013	09:10-10:50	11:05-12:05	10/02/2013	10:00-11:20	11:45-12:50
Event	09/11/2013	14:05-14:55	13:00-13:50	10/02/2013	13:50-14:50	15:35-16:15
Post-Event	09/12/2013	11:45-12:30	10:25-11:20	10/03/2013	10:30-11:15	11:45-12:30
One-Week	09/18/2013	13:15-14:15	11:20-12:40	10/09/2013	13:00-14:00	11:35-12:45

Physical data from the vertical profiles and grab sample results are discussed in the following sections. Vertical profiles included water temperature, DO, pH, and electrical conductance. Grab samples included hydrogen peroxide, nutrients, chlorophyll *a* and algae species, microcystin, and turbidity. Secchi measurements were made at each sampling event at each site.

4.2.1 Physical Data – Profiles: Water Temperature, Dissolved Oxygen, pH, and Electrical Conductivity

Water temperature, DO, pH, and electrical conductivity were measured at 0.5 m increments to 10.0 m or within 0.5 m of the bed (whichever was less) at each site during each sampling event. The monitoring results are summarized here for the September and October analysis periods, and all data are included graphically in the appendix.

In September, water temperature observations were consistent among all sites and indicated the presence of weak, intermittent stratification in near-surface water. Temperatures ranged from 20 to 23°C. In shallower water, vertical water temperatures typically varied throughout the water depth. In deeper profiles (e.g., non-treatment sites 1 and 2) the temperature profiles were roughly coincident below approximately 6 or 7 m. After one-week, the reservoir water was notably cooler and almost isothermal except the top 1 to 2 m. October temperatures were notably cooler, in the range of approximately 15 to 17.5°C. While there was some variability, overall the sites exhibited isothermal conditions in the top 10 m. The reservoir cooled 1 to 2°C during the one-week period that followed the October treatment event.

DO concentrations ranged from approximately 1 to 16 mg/L, with surface water samples illustrating higher concentrations than in deeper water. The lower DO values occurred in deeper water, due to a lack of mixing with the surface water caused by thermal stratification. Near-surface water concentrations were higher for event and post-event samples in both treatment and non-treatment sites. Water deeper than 5 to 6 m for the pre-event, event, and post-event sampling exhibited DO concentrations less than approximately 3 mg/L at all sites. A week later, DO concentrations were notably different, with fairly uniform concentrations top to bottom, ranging between roughly 6 and 10 mg/L depending on location. October DO concentrations were approximately uniform from top to bottom in the range of 5 to 7 mg/L, depending on location, depth, and sampling event.

During September, pH ranged from 8.5 to nearly 10 units with surface water experiencing higher values, consistent with the elevated pH response typically associated with the substantial primary production in the Klamath system. Deeper waters, where DO was below 3 mg/L, exhibited pH values below 8.5. Stratification was absent for the one-week sample, and pH was approximately uniformly distributed top to bottom at values less than 9. In October, under isothermal conditions, pH values varied between values of 8 and 9 among sites and were often fairly uniform top to bottom. One exception was within the treatment area for sites T1 and T3, where pH was higher in pre-event sampling than after treatment.

Electrical conductivity data were only collected during the September sampling period and ranged from 150 to 160 uS/cm in both the treatment and non-treatment sites. Surface water typically exhibited higher values, but bottom water occasionally exhibited elevated values. The one-week samples were generally lower. Overall, electrical conductivity was not affected by the treatment, as the conductivity at treatment sites responded similar to non-treatment sites during the study.

In summary, water temperature, DO, pH, and electrical conductivity, while exhibiting some variability, were largely consistent between treatment and non-treatment sites indicating that they were insensitive to the algaecide treatments.

4.2.2 Hydrogen peroxide

Samples were analyzed for hydrogen peroxide at the three treatment sites (T1, T2, and T3) before treatment and one-day after treatment in both September and October, consistent with permit requirements. Sampling results were non-detect for hydrogen peroxide in the pre-treatment and post-event sampling (Table 4).

Table 4. Summary of hydrogen peroxide analysis results (mg/L).

		Hydrogen Peroxide (mg/L)*			
		September		October	
Location	Depth (m)	Pre-Event (9/11/2013)	Post-Event (9/12/2013)	Pre-Event (10/2/2013)	Post-Event (10/3/2013)
T1	0.1	ND	ND	ND	ND
T2	0.1	ND	ND	ND	ND
T3	0.1	ND	ND	ND	ND

*ND means not detected at or above the reporting limit. Reporting limit is 1 mg/L.

4.2.3 Algal Response

Algae response to the treatment was measured by analysis of chlorophyll *a* concentrations and enumeration of cyanobacteria (BGA) species from the collected samples.

4.2.3.1 Chlorophyll a

In September, treatment area reductions were observed in average chlorophyll *a* concentrations in both the surface and integrated samples immediately after treatment and through the next week (Figure 6 and Table 5). Average chlorophyll *a* concentrations were reduced by 12 to 38 percent (from pre-event levels) during the day (event), were reduced by 72 to 81 percent through the next day (post-event), and were reduced by 82 to 91 percent after one week. By comparison, in the non-treatment area, average chlorophyll *a* concentrations increased, changed little, or declined over the course of one-week. Non-treatment surface samples exhibited over a two-fold increase (from pre-event levels) in chlorophyll *a* concentration during the day (event), a 40 percent increase (from pre-event levels) through the next day (post-event), and then a 46 percent reduction after one week. The integrated sample in the non-treatment area showed little change during the treatment day (event) or subsequent day (post-event), and then there was a 69 percent reduction after one week, which is less than the reduction observed in the average integrated treatment site sample.

In October, chlorophyll *a* concentrations were overall markedly lower than in September in both treatment and non-treatment areas. Treatment area surface and integrated samples exhibited a significant increase (from pre-event levels) in chlorophyll *a* concentration during the day (event) and through the next day (post-event). After one week, average chlorophyll *a* concentrations had returned to pre-event levels in both the surface and integrated samples. By comparison, in the non-treatment area, chlorophyll *a* concentrations remained relatively constant and low throughout the October sampling period.

Figure 6. Average chlorophyll *a* concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results (ppb), respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively. The error bar denotes the maximum and minimum observed concentration.

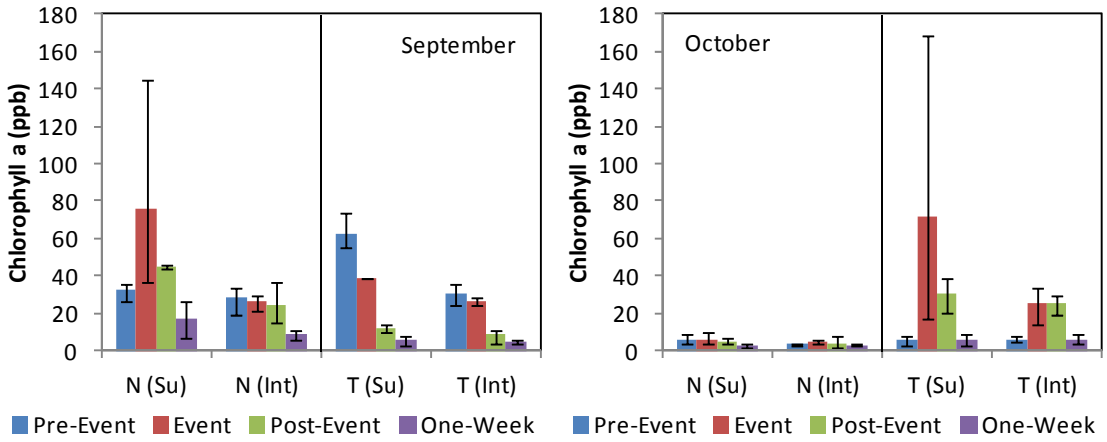


Table 5. Summary of average chlorophyll *a* response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

		Chlorophyll *a* (ppb)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	32	76	45	17	136%	40%	-46%
	N, Integrated	28	26	25	9	-6%	-12%	-69%
	T, Surface	62	39	12	6	-38%	-81%	-91%
	T, Integrated	30	27	8	5	-12%	-72%	-82%
October	N, Surface	6	6	5	3	4%	-8%	-50%
	N, Integrated	3	5	4	3	45%	26%	-7%
	T, Surface	5	72	31	6	1236%	465%	17%
	T, Integrated	6	25	26	6	321%	325%	-1%

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

4.2.3.2 Algae Species

Six types of algae species groups were identified in water samples taken from Long Gulch Cove: cyanobacteria (blue-green algae), chrysophyte (golden brown algae), cryptophyte, diatom, dinoflagellate, and green algae. In this study, algae species enumeration was limited to cyanobacteria because this is the algae species group of concern for toxin production. Algae species densities for *Microcystis aeruginosa* (MSAE) and *Aphanizomenon flos-aquae* (APFA), the most prevalent cyanobacteria, were analyzed (Figure 7 and Table 6 and Figure 8 and Table 7).

MSAE was present in all but one of the samples collected in September and October. In September, treatment area samples (surface and integrated) exhibited a general reduction in MSAE densities following treatment. MSAE densities varied from little change to a 38 percent increase (from pre-event levels) during the day (event). MSAE densities then were reduced by 28 to 68 percent through the next day (post-event), and were reduced by 66 to 89 percent after one week. By comparison, in the non-treatment area, MSAE densities increased, changed little, or declined less (than in the treatment area). MSAE densities in the non-treatment area samples (surface and integrated) varied from a 26 percent reduction (from pre-event levels) to an increase of over two-fold during the day (event). MSAE densities in the non-treatment area then increased by 65 to 88 percent (from pre-event levels) through the next day (post-event), and varied from a 43 percent reduction (from pre-event levels) to an increase of 91 percent after one week.

In October, the treatment area surface and integrated samples exhibited either an increase or little change (from pre-event levels) in MSAE densities during the day (event) and through the next day (post-event). However, after one week, MSAE densities in both the surface and integrated samples were reduced by about 80 percent from pre-event levels. By comparison, in the non-treatment area, MSAE densities remained relatively constant and low throughout the October sampling period.

APFA was present in 58 percent of the samples collected (60 samples out of 96 samples collected). The majority of the samples with APFA present were collected in September, and thus October conditions will not be discussed. In September, treatment area surface and integrated samples exhibited little change (from pre-event levels) in APHA densities during the day (event), but then declined by 15 to 73 percent through the next day and after one week. By comparison, in the non-treatment area, APFA densities declined after the pre-event sampling in nearly all cases (surface and integrated samples).

Figure 7. *Microcystis aeruginosa* (MSAE) concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results, respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively.

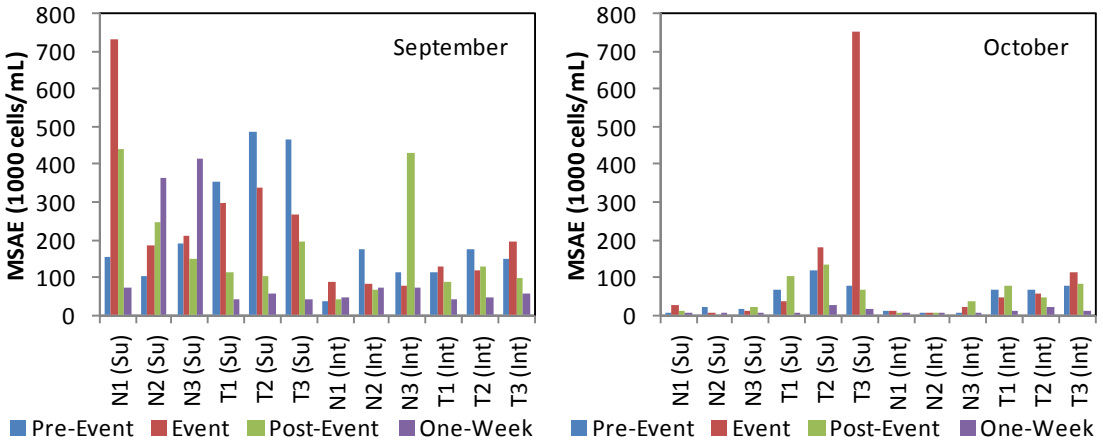


Table 6. Summary of average *Microcystis aeruginosa* (MSAE) response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

		MSAE (1,000 cells/mL)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	148	376	279	283	154%	88%	91%
	N, Integrated	109	81	180	62	-26%	65%	-43%
	T, Surface	434	301	137	47	-31%	-68%	-89%
	T, Integrated	146	149	105	49	2%	-28%	-66%
October	N, Surface	14	15	10	2	7%	-27%	-87%
	N, Integrated	7	13	16	1	77%	122%	-88%
	T, Surface	89	324	102	15	263%	14%	-83%
	T, Integrated	70	72	70	14	3%	-1%	-81%

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

Figure 8. *Aphanizomenon flos-aquae* (APFA) concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results, respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively. Note the scale change.

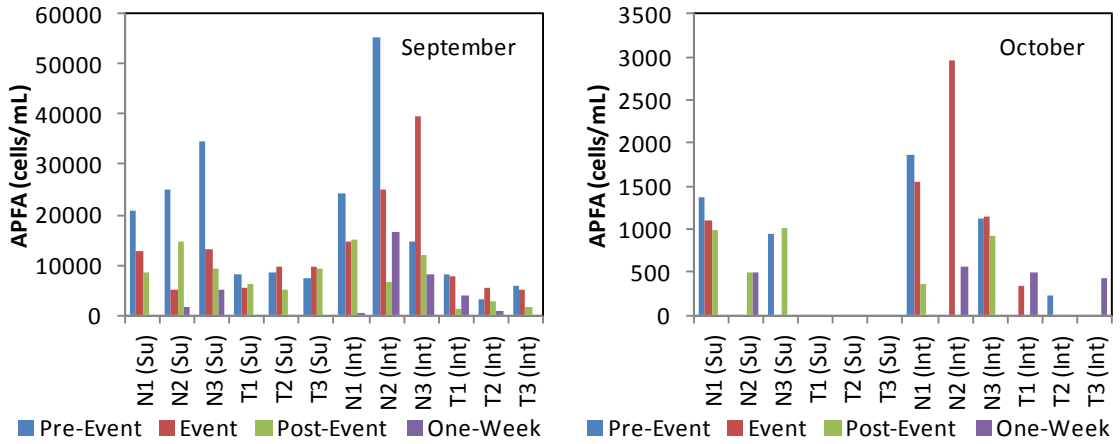


Table 7. Summary of average *Aphanizomenon flos-aquae* (APFA) response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

		APFA (cells/mL)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	26630	10320	10675	2201	-61%	-60%	-92%
	N, Integrated	31428	26409	11177	8376	-16%	-64%	-73%
	T, Surface	8102	8257	6860	-	2%	-15%	-
	T, Integrated	5650	5885	1908	1521	4%	-66%	-73%
October	N, Surface	207	-	-	-	-	-	-
	N, Integrated	-	545	2187	54	-	-	-
	T, Surface	557	731	1217	-	31%	118%	-
	T, Integrated	101	671	339	553	563%	235%	446%

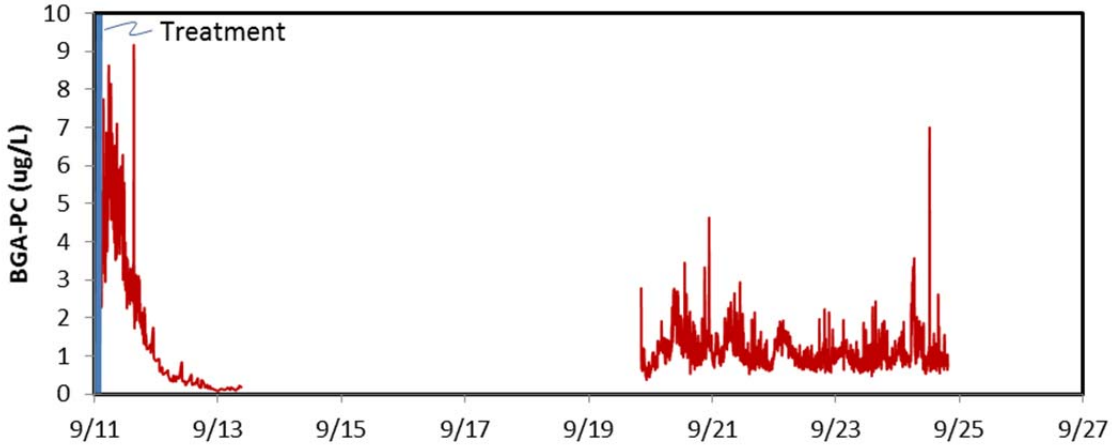
*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

The original study plan included the use of an automated vertical profiler containing a phycocyanin probe[2] to assess BGA conditions, but such information was not collected due to field equipment failure. However, phycocyanin information was collected using a non-automated fixed-depth probe that was available to be deployed for discrete periods during the study (Figure 9). While the resultant data do not provide vertical profiles (as originally planned), and there are data gaps, the information provides useful insight into post-treatment and longer-term responses. Specifically, the phycocyanin data indicate that BGA did not appear to immediately respond to treatment (e.g., event), but post-event

[2] The probe is used to detect (via florescence) the phycocyanin pigment found in cyanobacteria. Florescence readings can be correlated to quantitative data in order to provide concentration estimates.

levels (24 hours later) indicate a considerable reduction in BGA. Also, after approximately 10 days, there was a slight rebound – from near zero on September 13, 2013 to 1 to 2 ug/L on September 21, 2013. Subsequently, phycocyanin remains constant for several days.

Figure 9. Total BGA concentrations from the water quality sonde. The sonde was set at fixed depth of approximately 1.4 m from 14:10 on 09/11 to 08:20 on 09/12. The vertical profiler was operational from 08:45 on 09/12 to 21:00 on 09/13 and the sonde moved vertically through the water column (results are interpolated to be at 1.4 m). The sonde was not deployed from 21:00 on 09/13 through 08:20 on 09/20 due to mechanical difficulties with the profiler. The sonde was re-deployed at 08:25 on 09/20 through 07:35 on 09/25 at a depth of approximately 1.2 m.



4.2.3.3 Microcystin

In September, the average microcystin concentrations in both the surface and integrated treatment area samples declined by around 60 percent immediately after the treatment (event) (Figure 10 and Table 8). Treatment area surface samples exhibited a 49 percent reduction (from pre-event levels) in microcystin concentration through the next day (post-event), and then a 71 percent reduction after one week. On average treatment area integrated samples exhibited a nearly three-fold increase in microcystin concentration through the next day (post-event), and then a 61 percent reduction after one week. The spike in the post-event microcystin may have been due to heterogeneity in the sample area.

The results in October were variable and most likely influenced by notably lower pre-event concentrations. Outside of the treatment area, the concentrations of microcystin ranged from 2 to 4 ug/L. The curtain effectively isolated Long Gulch Cove and as a result, the pre-event concentrations at the treatment sites were higher (ranging from 9 to 26 ug/L). After algaecide was applied, microcystin concentration declined in two of the three sampling sites (T1 and T2), but increased in the third site (T3). After one-week, the average microcystin concentrations declined to below pre-event conditions in both the surface and integrated samples in the treatment area.

Figure 10. Microcystin concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results, respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively. The error bar denotes the maximum and minimum observed concentration. Note the scale change.

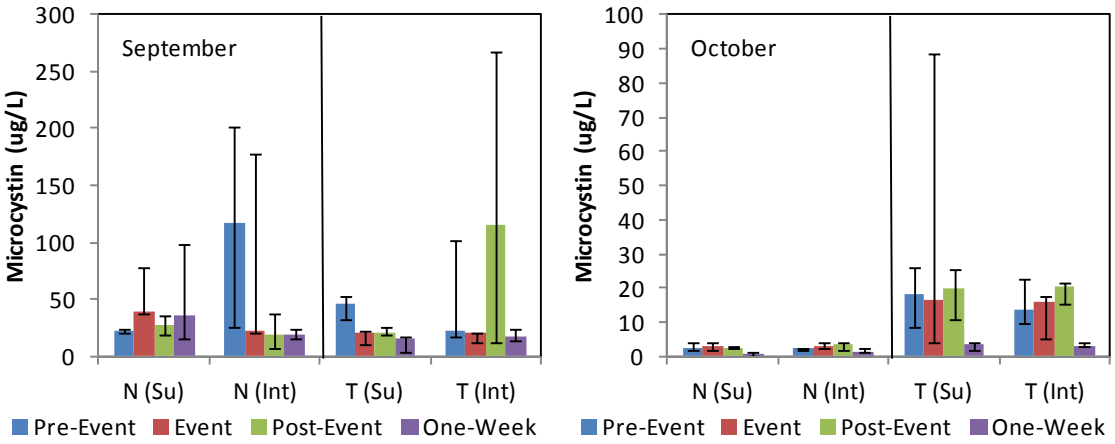


Table 8. Summary of average Microcystin response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

		Microcystin (ug/L)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	22.95	52.05	27.52	50.01	127%	20%	118%
	N, Integrated	115.40	73.68	21.59	19.69	-36%	-81%	-83%
	T, Surface	43.52	18.28	22.08	12.47	-58%	-49%	-71%
	T, Integrated	47.38	17.82	131.78	18.30	-62%	178%	-61%
October	N, Surface	2.92	3.03	2.62	0.83	4%	-10%	-72%
	N, Integrated	2.02	3.24	3.14	1.66	60%	55%	-18%
	T, Surface	17.54	36.37	18.73	3.28	107%	7%	-81%
	T, Integrated	15.50	12.87	19.12	3.34	-17%	23%	-78%

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

4.2.4 Turbidity

September turbidity for the treatment samples values generally showed a reduction in turbidity from pre-event to event, post-event, and one-week, while non-treatment sites showed mixed results with some samples increasing, some remaining approximately constant, and others decreasing in turbidity (Table 9). Systematic reductions from the pre-event to post-event and one-week samples suggest that treatment reduced turbidities. October sampling results were less evident with both treatment and non-treatment sites experiencing variable turbidity, where at times turbidity increased, remained approximately constant, or decreased.

Table 9. Summary of average turbidity measurements at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

		Turbidity (NTU)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	14.5	34.6	17.6	12.8	139	22	-11
	N, Integrated	8.9	7.3	7.5	7.2	-19	-16	-19
	T, Surface	36.6	23.5	16.9	9.5	-36	-54	-74
	T, Integrated	18.2	16.9	12.0	7.4	-7	-34	-59
October	N, Surface	2.1	2.4	2.3	2.0	14	11	-9
	N, Integrated	3.6	2.1	2.8	2.2	-43	-24	-41
	T, Surface	8.9	18.2	9.1	2.6	106	2	-70
	T, Integrated	6.8	10.7	7.6	2.0	58	12	-71

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

4.2.5 Secchi Disk

Secchi disk measurements in September ranged from 0.75 to 2.0 m. Readings were variable among sites and did not exhibit any clear trends between sites or sampling events (Table 10). October measurements ranged from 1.5 to 3.0 m, and similarly, treatment and non-treatment sites were variable without a clear trend.

Table 10. Summary of average Secchi disk measurements for both non-treatment ("Non-treatment") and treatment ("Treatment") area sites.

		Secchi Disk (m)				Percent Change from Pre-Event*		
	Sample Location	Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
Sep	Non-Treatment	1.4	0.9	1.0	1.5	-35	-29	6
	Treatment	0.9	0.8	1.0	1.7	-5	11	85
Oct	Non-Treatment	3.1	2.5	2.8	2.7	-17	-6	-11
	Treatment	1.9	1.6	1.8	2.7	-47	-42	-11

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

4.2.6 Nutrients

Grab samples were collected and analyzed for TN, NH4, NO2+NO3[3], TP, PO4, and DOC. In general, there were only small and/or inconsistent changes in the observed TN, NH4, TP, and DOC concentrations. See Appendix A for additional details. Only the NO2+NO3 and PO4 concentrations appeared consistently responsive to algaecide treatment.

September average NO3+NO2 concentrations increased in the treatment site for the event and post event samples, then decreased in the one-week sample (Figure 11 and Table 11). In the non-treatment site all values decreased. In October, the treatment site showed an increase in the post-event sample and at one-week. The non-treatment site only showed an increase in the one-week sample. Overall, October NO2+NO3 concentrations were higher than September concentrations and indicated only modest variability.

September average PO4 concentrations increased in the treatment site over the week (Figure 12 and Table 12). In the non-treatment site all concentrations were essentially unchanged except the surface site at one-week. In October, the treatment and non-treatment sites changed little through the week. Overall, PO4 concentrations in October were higher than September and indicted little variability.

[3] Grab samples were also analyzed for NO2, but all results were "non-detect." A "non-detect" concentration does not mean a concentration of zero, but rather that the concentrations were below the reporting limit. As such, NO2+NO3concentrations were analyzed instead.

Figure 11. Nitrate and Nitrite (NO3 + NO2) concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results, respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively. The error bar denotes the maximum and minimum observed concentration.

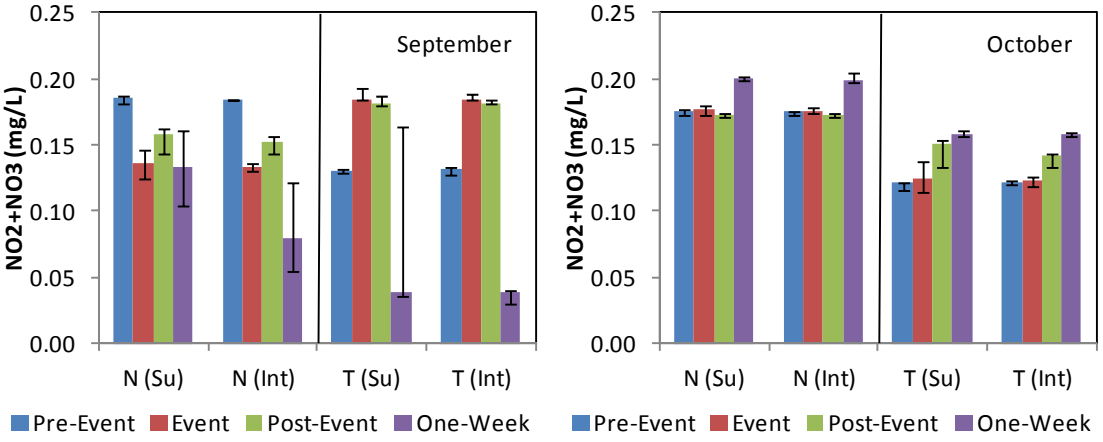


Table 11. Summary of average Nitrate and Nitrite (NO3+NO2) response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

	Sample Location	NO3+NO2 (ug/L)				Percent Change from Pre-Event*		
		Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	0.18	0.14	0.15	0.13	-26%	-16%	-28%
	N, Integrated	0.18	0.13	0.15	0.09	-28%	-18%	-54%
	T, Surface	0.13	0.19	0.18	0.08	43%	40%	-39%
	T, Integrated	0.13	0.19	0.18	0.04	42%	40%	-72%
October	N, Surface	0.18	0.18	0.17	0.20	1%	-1%	14%
	N, Integrated	0.17	0.18	0.17	0.20	1%	-1%	14%
	T, Surface	0.12	0.13	0.15	0.16	5%	22%	32%
	T, Integrated	0.12	0.12	0.14	0.16	1%	15%	30%

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

Figure 12. Orthophosphate (PO4) concentrations at all sampling locations and depths for September (left panel) and October (right panel). "N" and "T" denote non-treatment and treatment site sample results, respectively. "Su" and "Int" denote whether the samples were collected at the surface or were integrated, respectively. The error bar denotes the maximum and minimum observed concentration.

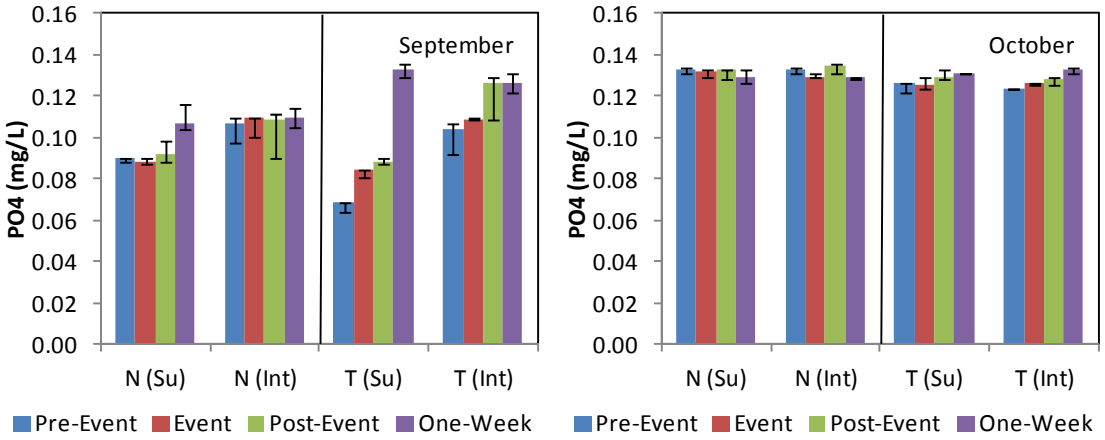


Table 12. Summary of average phosphate (PO4) response at the surface and integrated for both non-treatment ("N") and treatment ("T") area samples.

	Sample Location	PO4 (ug/L)				Percent Change from Pre-Event*		
		Pre-Event	Event	Post-Event	One-Week	To Event	To Post-Event	To One-Week
September	N, Surface	0.09	0.09	0.09	0.11	-1%	4%	21%
	N, Integrated	0.10	0.11	0.10	0.11	2%	-1%	5%
	T, Surface	0.07	0.08	0.09	0.13	23%	32%	97%
	T, Integrated	0.10	0.11	0.12	0.13	8%	21%	26%
October	N, Surface	0.13	0.13	0.13	0.13	-1%	-1%	-2%
	N, Integrated	0.13	0.13	0.13	0.13	-2%	1%	-3%
	T, Surface	0.12	0.13	0.13	0.13	1%	4%	5%
	T, Integrated	0.12	0.13	0.13	0.13	2%	3%	7%

*A positive percent change indicates concentrations increased between samplings and a negative percent change indicates a concentration decline between samplings.

5.0 DISCUSSION

As described above in the Introduction, the objectives of the 2013 in situ application of an environmentally safe algaecide in Long Gulch Cove focused on several technical objectives, including identifying the effects of peroxide-based algaecide application on algal response, microcystin levels, and nutrient concentrations. These technical objectives were successfully studied in the 2011 bench-top tests (Deas *et al*. 2012) and the 2012 pilot application study in Copco Cove (Deas *et al*. 2013). During the 2013 algaecide application study, the same technical objectives were studied in an isolated in-situ

reservoir setting. Described below are findings related to the two study periods (September and October), treatment and non-treatment sites; and the effects on water temperature, dissolved oxygen, pH, and electrical conductance, chlorophyll *a* and algal species, microcystin, and other constituents.

5.1 Study Events and Sampling Locations

The original study plan called for an initial, early treatment in Long Gulch Cove in 2013, while contemplating the potential for follow-up treatment(s) to manage algal conditions. However, the first treatment took place in September when extensive algal growth was already present. Several weeks later, based on available information (see Figure 9), a second, separate treatment was completed in early October. As noted previously, the September application employed different treatment levels at different depths to focus on the near-surface (7.5 ppm from 0 to 1.8 m) and deeper (2.5 ppm from 1.8 to 4 m) waters as distinct volumes. In October, only the upper 1.2 m of the water column was treated, and at a reduced application rate (2.5 ppm) based on lower algae concentrations.

Following the September treatment, atypical rainfall occurred in the area late in the month. Coupled with fall cooling in the reservoir and deepening of the epilimnion, differing meteorological conditions, reduced algal standing crop, and modified treatment levels, a direct comparison between the two treatments is unrealistic. As such, the results from the September application and the October applications are treated as two discrete events.

Another element considered in this study was the role of non-treatment sites and how the conditions outside the curtain may provide insight into conditions within the treatment area. The rationale for non-treatment site sampling was initially considered to provide a means to identify if conditions or trends between the sites were similar or different. While for the most part this did prove useful, a few confounding factors were identified that made direct comparison less informative. First, the non-treatment site was more exposed to and influenced by conditions in the main reservoir (e.g., wind and wave action) than the isolated treatment area. Second the depths at the non-treatment site (especially N1 and N2) were notably deeper than the treatment sites, and under isothermal conditions shallower waters were potentially able to mix with the deeper waters (>10 m) in the non-treatment locations. Finally, even though the non-treatment and treatment sites were influenced by different conditions, some parameters explored showed little deviation between the treatment and non-treatment sites (i.e., concentrations, measurement, or trends were similar regardless of sampling location). Because of the fundamental differences between the non-treatment and treatment locations, it was often difficult or not helpful to assess treatment efficacy based on comparisons between the treatment and non-treatment area (outside the curtain).

Both integrated and grab samples were included to explore conditions throughout the water column, as well as near-surface samples. In most cases integrated samples were similar to the near-surface water samples, with the exception of chlorophyll *a*, algae species, and microcystin. NO2+NO3 and PO4 exhibited systematic differences in the

treatment area after treatment. All other constituents illustrated minor or infrequent deviations between integrated and near-surface samples.

5.2 Water Temperature, Dissolved Oxygen, pH, and Electrical Conductance

The key finding from the profiles of physical parameters was that weak, intermittent stratification within the epilimnion had an effect on some water quality constituents. DO, pH, and electrical conductance all responded to thermal stratification in the pre-event, event, and post-event sampling. A few days after the September treatment, the weak, intermittent stratification broke down, and most parameters at the treatment and non-treatment sites were fairly uniform from the water surface to the reservoir bed (or deepest observation point). This isothermal condition existed during the October event.

DO values in the water column were affected by the stratified conditions. When stratification was present, there was a lack of mixing between surface layers and deeper water. Higher DO concentrations occurred in the surface layers, where primary production was higher and where oxygen exchange could occur across the air-water interface. Lower DO concentrations occurred in the deeper water, where stratification isolated the region from reaeration at the lake surface and also where algal concentrations (and primary production) were lower. Because treatment and non-treatment sites both exhibited increased DO concentrations following treatment in near surface (0 to 2 m) waters, no real impact on DO was apparent. Day to day variability in photosynthesis due to variation in primary production, area heterogeneity, meteorological conditions and other factors also played a role in DO dynamics in the near-surface waters of the reservoir. Field data suggest that treatment did not substantially affect DO. October DO conditions were largely uniform from top to bottom at all sites and similar among sites.

Treatment had no noticeable effect on pH and electrical conductance, which is consistent with manufacturer guidelines (V. Choppakatla pers. comm.).

5.3 Hydrogen peroxide

Hydrogen peroxide, the active ingredient in GreenClean Liquid, was not detected within the treatment area after application. This finding is consistent with manufacturer application rate guidelines and the chemistry of product, which reacts rapidly after application (seconds to minutes), degrading to water and oxygen.

5.4 Chlorophyll *a*

The longer-term (one-week) response of chlorophyll *a* concentrations to the application of algaecide was fairly consistent in the individual September and October events. In September, on average, chlorophyll *a* concentrations were reduced by 63 percent after algaecide treatment, thus indicating that GreenClean Liquid was able to effectively damage or kill algal cells in the reservoir. Chlorophyll *a* concentrations remained substantially below pre-event levels throughout the September study period. This is consistent with input provided by both the manufacturer and applicator that maximum reductions typically occur after a day and up to a week following application.

In October, surface and integrated sample chlorophyll *a* concentrations increased after treatment before returning to pre-event concentrations within one week. The heterogeneous nature of the algae in the treatment and non-treatment area, cooler water temperatures, and different meteorology contributed to mixed results of the October sampling. The low application dose and shallow depth, combined with isothermal conditions may have led to dilution during application, with shallow waters readily mixing with deeper waters (in the absence of a temperature (density) gradient) during treatment. During the October treatment, field crews noted the "patchy" nature of surface accumulations, and that this may reduce the representativeness of the sampling sites within the sampling area. Chlorophyll *a* concentrations were generally higher in the treatment area than in the non-treatment sites. A possible explanation is that the enclosure provided a condition more conducive for growth than non-treatment (main reservoir) sites at this time of year, but more data would be required to verify this condition.

Pre-event concentrations were markedly different between the two discrete events in September and October, where chlorophyll *a* concentrations ranged from 24 to 74 ppb in September, but only ranged from 3 to 8 ppb in October. This general reduction in chlorophyll *a* concentrations between September and October was also observed in the non-treatment site samples, indicating that natural processes at the reservoir scale (e.g., fall senescence of seasonal phytoplankton growth, transitioning to isothermal conditions, cooler water temperatures), also led to chlorophyll *a* concentrations diminishing between the months of September and October.

5.5 Algae Species

In terms of MSAE, surface water concentrations in September exhibited a noticeable and consistent reduction after the application of GreenClean Liquid. All of the September treatment site surface samples exhibited a reduction in MSAE after treatment and one week later (15 to 91 percent). This is especially noteworthy since the near-surface samples represent waters that pose the greatest potential health risk. Most of the September integrated samples also exhibited a reduction in MSAE concentrations. These reductions in MSAE concentrations at the treatment sites were in contrast to the concentration pattern exhibited at the non-treatment sites, where surface samples generally exhibited an increase in MSAE concentrations in September, while the integrated samples concentrations were variable. These results are also consistent with the chlorophyll *a* results for the treatment and non-treatment site.

The results in October were more variable in the short-term (first 24-hours), but all sites (including the non-treatment site) and samples exhibited a reduction after one week (69 to 89 percent). It is not clear if the changes in the October MSAE concentrations were due to the application of algaecide or natural processes within the water body. October cell counts were lower and though results were not conclusive, MSAE was more prevalent in the treatment area than the non-treatment area, consistent with the chlorophyll *a* data as discussed, above.

APHA did not follow the trends observed in MSAE. Rather, treatment conditions were variable, but did exhibit some similar patterns to the non-treatment sites, mainly regarding the reductions in post-event and one-week sample concentrations. However, because of the lack of APHA in samples and the low concentrations, no conclusions can be drawn regarding the treatment efficacy for APHA. Unlike MSAE, APHA was not found in higher concentrations in the treatment area compared to the non-treatment area.

5.6 Microcystin

Microcystin response to GreenClean Liquid was variable among the two sampling periods. In general, September surface samples exhibited a reduction in concentration after application. Most of the integrated samples also experienced a reduction in total microcystin concentration after treatment, but two of the samples exhibited notable increases one-day later (post-event). This increase in microcystin concentrations one day after treatment was most likely due to heterogeneity in the sampling area, or other factors. One week after treatment the levels were lower than pre-event concentrations.

October results were variable and may be influenced by lower pre-event concentrations. Visual observations in the study area indicated notable heterogeneity in surface accumulations of algae – areas with little algae and other areas where wind and other conditions led to notable algae. While such variability is common for algae, these low populations appear to have led to more contrast. Modest winds, light conditions, small scale stratification, and other factors all may contribute to patchy distribution of algae within the study area.

While the 2012 study identified a potentially strong relationship between treatment and microcystin reductions, the 2013 results are less clear. Differences between the two studies may include the longer sampling period, larger area, larger volume, greater depths, different treatment approaches, and other factors. While 2013 results suggest microcystin concentrations are reduced following treatment (at the one-week time frame), additional studies would aid in understanding the implications at the scale of the Long Gulch Cove area.

5.7 Secchi Depth and Turbidity

Secchi depth did not indicate any conclusive response to treatment. Secchi depths in October were larger than September, but this is probably due to the overall reduction in primary production as summer transitions to fall.

Turbidity values decreases consistently in treatment sites in both months, suggesting that algaecide application had a direct effect. October values were lower than September values, due to the same reasons as mentioned previously for Secchi depth. While Secchi depth information was not conclusive, turbidity is a more quantitative measure and produced clear trends.

5.8 Nutrients

Overall, treatment implications for total and inorganic nitrogen and phosphorus, and dissolved organic carbon appear to be limited to:

- Short duration increases in NO2+NO3 (September) where event and post event samples showed increases (one-week samples showed decreases consistent with the non-treatment sites).

- Increases in PO4 where event, post-event, and one-week samples illustrate larger increases in the treatment samples than in the non-treatment samples.

Nutrient sampling for TN, NH4, TP, and DOC do not provide a conclusive pattern of either a notable increase or decrease, although TN and TP values did reflect algae patterns, due to the organic matter fraction included in algal communities. The increase in NO2+NO3 and PO4 would be attributed be due to cell lysing during treatment, reduced algal uptake due to reductions in algal standing crop following treatment, and natural variability within the treatment area.

5.9 Summary

Important findings of this exploratory application of hydrogen peroxide-based algaecide indicate that chlorophyll *a*, MSAE counts, NO2+NO3, PO4, and turbidity were affected by treatment. Algae conditions, as measured by chlorophyll *a*, MSAE and microcystin were generally reduced after treatment, as was turbidity. The inorganic forms, NO2+NO3 and PO4, increased after treatment. All of these conditions were present at the end of the sampling week. These findings are consistent with previous work completed for PacifiCorp. Many parameters were found to be largely insensitive to treatment, including temperature, dissolved oxygen, pH, electrical conductance, TN, TP, NH4, DOC, and Secchi depth. Hydrogen peroxide, the active ingredient in GreenClean Liquid was absent in samples collected after application. While temperature was not observably impacted by the treatment, the role of stratification in design and application of algaecide, as well as the overall water quality response of reservoir waters, was an important finding.

The integrated sampling to 6 meters (or to within 0.5 m of the bed where shallower waters were encountered), provided a means to assess if surface waters were notably different than deeper waters. While a useful experimental element, the need for such samples may not be as important as originally considered.

6.0 CONCLUSION AND RECOMMENDATIONS

The 2013 study of the application of environmentally safe algaecide in Long Gulch Cove in Iron Gate reservoir was designed based on information developed from previous bench-scale studies conducted in 2008, 2009, and 2011 (Deas *et al*. 2009; 2012) and the pilot study in Copco Cove (Deas *et al*. 2013). The 2013 pilot test application in Iron Gate reservoir addressed several objectives:

- Installation of a curtain system in a reservoir to enclose a volume of water for treatment.
- Evaluation of the effectiveness of algaecide within this enclosed reservoir area in reducing algal cells by observing the response of chlorophyll *a,* algae species counts, and other physical and chemical constituents to the algaecide application over a period of up to one week.
- Determination of the impact of algaecide application on microcystin concentrations.
- Definition of the necessary steps and activities associated multiple algaecide applications.

Overall, the results of the 2013 pilot application study indicated that GreenClean Liquid is effective in reducing blue-green algae in a confined reservoir environment and may potentially reduce microcystin concentrations. Reductions in chlorophyll *a* and algal species constituents showed that the application of the hydrogen peroxide-based algaecide was effective in killing algal cells and reducing their overall levels. In addition, algaecide treatment led to modest increases in NO3+NO2 and PO4 concentrations immediately after treatment. These increases were assumed to be a consequence of reduction in algal uptake and release of inorganic nutrients from algal cell death and lysis. However, other nutrient concentrations (e.g., ammonium and dissolved organic carbon) were generally unchanged. GreenClean Liquid application was also shown to have the potential to reduce microcystin levels within the treated area of the reservoir through several possible mechanisms.

These findings are largely based on the September period when algal presence was notable (i.e., chlorophyll *a* and algae species counts were high). While October results provide additional useful insight, overall reservoir conditions lead to low algal concentrations, and monitoring results were less conclusive. Overall, the results of the 2013 application study indicated that GreenClean Liquid was effective in reducing blue-green algae in the reservoir environment.

Based on the 2013 findings, as well as previous algaecide experiments, recommendations for future work included:

- **Start application earlier in the year within the isolated area to assess algal management through the summer season.** The 2013 study occurred in early-September and early-October. By early-September the algae standing crop within Iron Gate reservoir had already developed to a level at which a medium to high algaecide application rate was recommended by the applicator. By mid-October, the standing algae crop had declined and a low dosage was applied. An earlier application of algaecide would allow for an assessment of the effectiveness of algaecide in preventing the development of a large standing crop that adversely affects reservoir water quality conditions and thus requires higher algaecide application rates.
- **Continue to refine sampling approach and constituents examined**, including a wider use of phycocyanin probes for more detailed spatial monitoring (e.g., vertical profiling and/or multiple locations at a fixed depth or depths). For potential future

studies, a balance of resources and appropriate level of monitoring should be identified to (a) meet applicable regulatory requirements, and (b) effectively characterize conditions in treated areas. It is recommended that monitoring focuses on chlorophyll *a*, algae species, microcystin, as well as regulatory requirements. At this time it appears that performing surface and integrated samples is not necessary and that a single grab sample collected at 3 ft (0.9 m) depth (consistent with applicable permit requirements) would be sufficient to evaluate effectiveness. It is recommended that TN, TP, NH4, and DOC grab sampling be discontinued.

While a goal in 2013, equipment malfunction precluded the implementation of a vertical profiler in the treatment area. The placement of a vertical profiler in the treatment area is recommended. The use of a phycocyanin probe when completing vertical profiles or to sample spatially in the treatment area would be useful to characterize BGA distributions.

- **Development of a plan that balances resources with an appropriate level of monitoring during future algaecide applications**. The monitoring completed in the 2013 study identified that the barrier was sufficient to isolate a portion of Long Gulch Cove from the rest of Iron Gate reservoir. The isolated portion had different water quality conditions prior to treatment, such that the non-treatment sites could no longer be considered "control" samples. The two sets of samples could be qualitatively compared to determine if similar trends/patterns were observed, but a direct comparison was not necessarily applicable given the differences between the sites. With the knowledge gained from the various experiments over the past several years, sampling would be better focused on the key constituents within the treatment area. The non-treatment area has the potential to exhibit higher variability, is notably deeper, and is difficult to compare to the treatment area. Focusing on the treatment area with a goal of maintaining reduced algal concentrations would provide a more direct means of assessing the efficacy of the algaecide application and management options to reduce harmful algae blooms in specific (localized) areas of the lake (e.g., high use areas).
- **Focus evaluation efforts on the effectiveness of algaecide in reducing algal cells by observing the response of chlorophyll *a* and cyanobacteria species to the algaecide application**. Further studies should continue to monitor cyanobacteria through selected sampling, to explore the relationship between microcystin concentrations and algal species presence. This will allow for a better understanding of the relationships between algal species composition and microcystin concentrations.

This study was the second application of hydrogen peroxide-based, environmentally safe algaecide to Klamath River reservoirs and the first to an enclosed volume of water within a reservoir. While the application was completed with critical knowledge gained from the 2012 application at Copco reservoir, the application to a confined cove of the lake and different application rates at different depths were new features. Sampling methods were modified to capture system response to treatment and a wide range of parameters were identified for analysis. Algaecide applications were also carried out at two times of year that presented notably different conditions. The early September application occurred

with stratification present and a notable standing crop of algae. This application, which was largely successful in reducing chlorophyll *a* and microcystis concentrations, yielded information consistent with managing a more typical algae condition in the reservoir. The October application occurred during isothermal conditions in the study area and low standing crop of algae. For many constituents, there was little change among the treatment and non-treatment areas, as well as between the integrated and near-surface grab samples. However, there were several constituents that provided clear trends with regards to treatment efficacy.

The manufacturer and applicators identified the principal constituent used to measure efficacy is chlorophyll *a*. Other associated constituents that effectively illustrated a treatment response were related: algae counts for MSAE, microcystin, and turbidity. Treatment effects may not be identified for a day or more (up to a week after treatment), and for these constituents the one-week sample nearly always showed a reduction from pre-event sampling. The September treatment may be termed more "successful" because results indicated clear trends in algae reductions. October results were more variable, but there were also confounding factors including low standing crop, different algaecide application rates and depths, isothermal conditions, variable meteorological conditions, and other factors.

The study provided an opportunity to explore an enclosed reservoir area and treating that volume over an extended, albeit short, period. Different treatment approaches were applied, monitoring strategies developed and tested, and a range of meteorological and reservoir conditions exhibited. From this successful experimental effort, several lessons were learned and recommendations for future studies are outlined previously.

Overall, the 2013 treatment study in Long Gulch Cove demonstrated that algaecide application effectively reduced algal concentrations, reduced surface water microcystin concentrations, and reduced algal biomass (as measured by chlorophyll *a*). However, natural lake processes, such as destratification and cooling, and the addition of the barrier to isolate a portion of the cove affected chlorophyll *a*, algae densities, microcystin, and other constituent concentrations during the study periods. Overall, short-term reductions in microcystin, cholorophyll *a*, and algae concentrations indicate that a hydrogen peroxide-based, environmentally-safe algaecide could potentially be a useful management tool to reduce algal production and associated algal toxins in selected areas.

7.0 REFERENCES

Antoniou, M.G., A.A. de la Cruz, and D.D. Dionysiou. 2005. "Cyanotoxins: New Generation of Water Contaminants." *J Environ Eng*. **131**(9): 1239–1243.

Barrington, D.J., and A. Ghadouani. 2008. "Application of Hydrogen Peroxide for the Removal of Toxic Cyanobacteria and Other Phytoplankton from Wastewater." *Environ Sci Technol.* **42**: 8916–8921.

Barrington, D.J., A. Ghadouani, and G.N. Ivey. 2011. "Environmental Factors and the Application of Hydrogen Peroxide for the Removal of Toxic Cyanobacteria from Waste Stabilization Ponds." *J Environ Eng*. **137**(10): 952-960.

Barrington, D.J., E.S. Reichwaldt, and A. Ghadouani. 2013. "The Use of Hydrogen Peroxide to Remove Cyanobacteria and Microcystins from Waste Stabilization Ponds and Hypereutropic Systems." *Ecol Eng.* **50**: 86-94**.**

Berman, T. 1997. "Dissolved Organic Nitrogen Utilization by an Aphanizomenon Bloom in Lake Kinneret." *J. Plankton Res*. **19**: 577-586.

Berman, T. and S. Chava. 1999. "Algal Growth on Organic Compounds as Nitrogen Sources." *J. Plankton Res.* **21**:1423-1437.

BioSafe Systems L.L.C. (Biosafe). 2012. GreenClean® Liquid Broad Spectrum Algaecide/Bactericide – Specimen Label. (Available online at: http://www.biosafesystems.com/assets/greencleanpro-specimen-label.pdf)

Carmichael, W.W., C. Drapeau, and D.M. Anderson. 2000. "Harvesting of *Aphanizomenon flos-aquae* ralfs ex Born and flah. Var. *flosaquae* (Cyanobacteria) from Klamath Lake for Human Dietary Use." *J Appl Phycol.* **12**: 585–595.

Cooke, G.D., E.B. Welch, S. Peterson, and S.A. Nichols. 2005. *Restoration and Management of Lakes and Reservoirs*. Third Edition. CRC Press.

Cooper, W.J., and R.G. Zika. 1983. "Photochemical Formation of H2O2 in Surface and Ground Waters Exposed to Sunlight." *Science.* **220**: 711–712.

Deas, M.L., J.C. Vaughn, and S.K. Tanaka. 2009. *Algaecide Pilot Study: Copco Reservoir 2008.* Prepared for PacifiCorp. November 30.[NALMS] North American Lake Management Society (2007). "NALMS – Blue Green Algae." http://www.nalms.org/Resources/BlueGreenInitiative/Overview.htm

Deas, M.L., S.K. Tanaka, E. Limanto, and E. Miao. 2012. Pilot Testing of Environmentally-Safe Algaecide on Copco Reservoir Water – 2011 Study Results. Prepared for PacifiCorp. December 10, 2012. 46 pp.

Diffey, D.L. 2002. "Sources and measurement of ultraviolet radiation." *Methods*. 28, 4-13.

Ding, Y., N. Gan, J. Li, B. Sedmak, and L. Song. 2012. "Hydrogen Peroxide Induces Apoptotic-like Cell Death in *Microcystis aeruginosa* (Chroococcales, Cyanobacteria) in a Dose-dependent Manner." *Phycologia* **51**: 567–575.

Doane, T.A., and W.R. Horwath. 2003. "Spectrophotometric Determination of Nitrate with a Single Reagent." *Analytical Letters*. **36**(12):2713-2722. Marcel Dekker Publishing.

Drábková, M., W. Admiraal, and B. Marsálek. 2007. "Combined Exposure to Hydrogen Peroxide and Light–selective Effects on Cyanobacteria, Green Algae, and Diatoms." *Environ Sci Technol.* **41**: 309–314.

Ellis, B. K., and J.A. Stanford. 1982. "Comparative Photoheterotrophy, Chemoheterotrophy, and Photolithotrophy in a Eutrophic Reservoir and an Oligotrophic Lake." *Limnol. Oceanogr*. **27**:440-454.

Engelsen, O. 2005. FastRT - Fast simulations of downward UV doses, indices and irradiances at the Earth's surface. Norwegian Institute for Air Research, N-9296, Tromsø, Norway. (available at: http://nadir.nilu.no/~olaeng/fastrt/fastrt.html).

Eisenberg, G. 1943. "Colorimetric Determination of Hydrogen Peroxide." *Ind. Eng. Chem. Anal. Ed.*, 1943, **15**(5): 327–328.

Environmental Protection Agency. 2002, Sodium Carbonate Peroxyhydrate (128860) Fact Sheet. Office of Pesticide Programs, EPA, 1200 Pennsylvania Ave., Washington, D.C.

Fan, Jiajia, P. Hobson, L. Ho, R. Daly, and J. Brookes. 2014. "The Effects of Various Control and Water Treatment Processes on the Membrane Integrity and Toxin Fate of Cyanobacteria." *J Hazard Mater*. 264: 313-322.

Horne, A.J., and C.R. Goldman. 1994. *Limnology*, Second Edition. McGraw-Hill, Inc. New York, NY.

Kenefick, S.L., S.E. Hrudey, H.G. Peterson, and E.E. Prepas. 1993. "Toxin Release from *Microcystis aeruginosa* After Chemical Treatment." *Water Sci. Technol*. **27**(3–4): 433–440.

Kirkwood, A. E., C. Nalewajko, and R.R. Fulthorpe. 2003. "Physiological Characteristics of Cyanobacteria in Pulp and Paper Waste–Treatment Systems." *J Appl Phycol*. **15**:325-335.

Knox, K. 2009. Peracetic Acid Petition. BioSafe Systems LLC. Docket No. AMS-TM-09-0014.

Larose, R., Fisher, P., Austen, E., and Choppakatla. V. 2008. "Water Treatment Series: Activated Peracids Can Treat Water." *Greenhouse Management and Production* **28**(11): 14-19.

Lawton, L.A., Robertson, P.K.J., Cornish, B.J.P.A., Jaspars, M., 1999. Detoxification of microcystins (cyanobacterial hepatotoxins) using TiO2 photocatalytic oxidation. Environmental Science and Technology 33 (5).: 771-775.

Li, R., W.W. Carmichael, and P. Pereira. 2003. "Morphological and 16S Gene Evidence for Reclassification of the Paralytic Shellfish Toxin Producing *Aphanizomenon flos-aquae* LMECYA 31 as *Aphanizomenon issatschenkoi* (Cyanophyceae)." *J Appl Phycol.* **39**: 814–818.

Liu, I., Lawton, L.A., Cornish, B., Robertson, P.K.J., 2002. Mechanistic and toxicity studies of the photocatalytic oxidation of microcystin□LR. Journal of Photochemistry and Photobiology A, Chemistry 148 (13), 349-354.

Matthijs, H.C.P., P.M. Visser, B. Reeze, J. Meeuse, P.C. Slot, G. Wijn, R. Talens, and J. Huisman. 2011. "Selective Suppression of Harmful Cyanobacteria in an Entire Lake with Hydrogen Peroxide." *Water Res*. **46**: 1460-1472.

McElhiney, J., L.A. Lawton. 2005. "Detection of the Cyanobacterial Hepatotoxins Microcystins." *Toxicol. Appl. Pharmacol*. **203**(3): 219–230.

Olvera-Ramírez, R., C. Centeno-Ramos and F. Martínez-Jerónimo. 2010. "Toxic Effects of *Pseudanabaena tenuis* (Cyanobacteria) on the Cladocerans Daphnia magna and Ceriodaphnia dubia." *Hidrobiológica* **20** (3): 203-212.

Oudra, B., M. Loudiki, B. Sbiyyaa, R. Martins, V. Vasconcelos, and N. Namikoshi. 2001. "Isolation, Characterization and Quantification of Microcystins (heptapeptides hepatotoxins) in *Microcystis aeruginosa* Dominated Bloom of Lalla Takerkoust Lake/Reservoir (Morocco)." *Toxicon.* **39**: 1375-1381.

Oudra, B., M. Loudiki, V. Vasconcelos, B. Sabour, B. Sbiyyaa, K. Oufdou, and N. Mezrioui. 2002. "Detection and Quantification of Microcystins from Cyanobacteria Strains Isolated from Reservoirs and Ponds in Morocco." *Environ Toxicol*. **17**: 32-39.

Paerl, H.W. 2008. "Nutrient and Other Environmental Controls of Harmful Cyanobacterial Blooms Along the Freshwater-Marine Continuum." *Adv Exp Med Biol.* **619**: 216–241.

Qian H, S. Yu, Z. Sun, X. Xie, W. Liu, and Z. Fu. 2010. "Effects of Copper Sulfate, Hydrogen Peroxide and N-phenyl-2-naphthylamine on Oxidative Stress and the Expression of Genes Involved Photosynthesis and Microcystin Disposition in *Microcystis aeruginosa*." *Aquat Toxicol.* **99**: 405–412.

Qian, H., B. Hu, S. Yu, X. Pan, and T. Wu. 2012. "The Effects of Hydrogen Peroxide on the Circadian Rhythms of Microcystis aeruginosa." PLoS ONE **7**(3): e33347. doi:10.1371/journal.pone.0033347.

Raymond, R. 2008. Results of 2007 Phytoplankton Sampling in the Klamath River and Klamath Hydroelectric Project (FERC 2082). Prepared for PacifiCorp. December 12.

Raymond, R. 2009. Phytoplankton Species and Abundance Observed During 2008 in the Vicinity of the Klamath Hydroelectric Project. Prepared for PacifiCorp. September.

Raymond, R. 2010. Phytoplankton Species and Abundance Observed During 2009 in the Vicinity of the Klamath Hydroelectric Project. Prepared for PacifiCorp. July.

Reichwaldt, E.S., L. Zheng, D.J. Barrington, and A. Ghadouasni. 2012. "Acute Toxicological Response of *Daphnia* and *Moina* to Hydrogen Peroxide." *J. Environ. Eng.* **138**: 607-611.

Rodrıguez, E., Majado, M.E., Meriluoto, J., Acero, J.L., 2007. Oxidation of microcystins by permanganate: reaction kinetics and implications for water treatment. Water Research 41 (1), 102-110.

Rodrıguez, E.M., Acero, J.L., Spoof, L., Meriluoto, J., 2008. Oxidation of MC□LR and □RR with chlorine and potassium permanganate: toxicity of the reaction products. Water Research 42 (67), 1744-1752.

Ross, C., L. Santiago-Vazquez, and V. Paul. 2006. "Toxin Release in Response to Oxidative Stress and Programmed Cell Death in the Cyanobacterium *Microcystis aeruginosa*." *Aquat Toxicol.* **78**: 66–73.

Saker, M.L., A. D. Jungblut, B.A. Neilan, D.F.K. Rawn, and V.M. Vasconcelos. 2005. "Detection of Microcystin Synthetase Genes in Health Food Supplements Containing the Freshwater Cyanobacterium *Aphanizomenon flos-aquae*." *Toxicon* **46**: 555–562.

Samuilov, V. D., K.N. Timofeev, S.V. Sinitsyn, and D.V. Bezryadnov. 2004. "H2O2-Induced Inhibition of Photosynthetic O2 Evolution by Anabaena variabilis Cells." *Biochemistry.* **69**(8): 926–933.

Scully, N.M., D.R.S. Lean, D.J. McQueen, and W.J. Cooper. 1995. "Photochemical Formation of Hydrogen Peroxide in Lakes: Effects of Dissolved Organic Carbon and Ultraviolet Radiation." *Can B Fish Aquat Sci*. **52**(12): 2675-2681.

Scully, N.M., D.J. McQueen, W.J. Cooper, and D.R.S. Lean. 1996. "Hydrogen Peroxide Formation: The Interaction of Ultraviolet Radiation and Dissolved Organic Carbon in Lake Waters Along a 43-75 degree N Gradient." *Limnol Oceanogr.* **41**(3): 540-548.

Skurlatov, Y.I., and L.S. Ernestova LS. 1998. "The Impact of Human Activities on Freshwater Aquatic Systems." *Acta Hydrochim. Hydrobiol.* **26**(1): 5–12.

Stevenson, R J., M.L. Bothwell and R.L. Lowe. 1996. *Algal Ecology: Freshwater Benthic Ecosystems*. Academic Press, Inc. San Diego, California. 40-41 pp.

Svrcek, C., and D.W. Smith. 2004. "Cyanobacteria Toxins and the Current State of Knowledge on Water Treatment Options: a Review." *J Environ Eng*. **3**(3), 155–185.

United States Department of Interior, Bureau of Reclamation, Environmental Monitoring Branch (USBR). 2009. *Standard Operating Procedures for Quality Assurance*. U.S. Department of the Interior, Bureau of Reclamation, Mid-Pacific Region.

United States Environmental Protection Agency (EPA). 1978. National Eutrophication Survey Report on Iron Gate Reservoir, Siskiyou County, CA. EPA Region IX, Working Paper No. 749.

United States Environmental Protection Agency (EPA). 2012. "Pesticides: Regulating Pesticides." September. <http://www.epa.gov/oppbppd1/biopesticides/index.htm>

Vincent W. F. and C.R. Goldman. 1980. "Evidence for Algal Heterotrophy in Lake Tahoe, California, Nevada." *Limnol Oceanogr*. **25**:89-99.

Wagner, K.J. 2004. "The Practical Guide to Lake Management in Massachusetts: A Companion to the Final Generic Environmental Impact Report on Eutrophication and Aquatic Plant Management in Massachusetts." Commonwealth of Massachusetts Executive Office of Environmental Affairs.

Watercourse Engineering, Inc. (Watercourse). 2013. 2012 Localized Treatment of Copco Cove in Copco Reservoir Using Environmentally Safe Algaecide Prepared for PacifiCorp Energy, Portland OR. July. 57 pp.

World Health Organization (WHO). 1999. "Toxic Cyanobacteria in Water: A guide to their public health consequences, monitoring and management." Chapter 3 <http://www.who.int/water_sanitation_health/resourcesquality/toxcyanobacteria.pdf>

World Health Organization (WHO). 2003. Cyanobacterial toxins: Microcystin-LR in Drinking-water - Pre-Event document for development of WHO Guidelines for Drinking-water Quality. WHO/SDE/WSH/03.04/57.

World Health Organization (WHO). 2007. Draft Second Amendment on microcystin treatment for inclusion in the Guidelines for Drinking Water. http://www.who.int/water_sanitation_health/dwq/chemicals/microcystin_sections.pdf

Zhou, S. Y. Shao, N. Gao, Y. Deng, J. Qiao, H. Ou, and J. Deng. 2013. "Effects of Different Algaecides on the Photosynthetic Capacity, Cell Integrity and Microcystin-LR Release of *Microcystis aeruginosa*." *Sci Total Environ.* **463-464**: 111-119.

Znachor, P. and J. Nedoma. 2009. “Importance of Dissolved Organic Carbon for Phytoplankton Nutrition in a Eutrophic Reservoir.” *J Plankton Res*. **32** (3): 367-376.

Personal Communications

Jeff Kline, BioSafe Systems, LLC. April 29, 2010; July 19, 2012; October 9, 2012; January 28, 2013.

Vijay Kumar Choppakatla, PhD, BioSafe Systems, LLC. July 19, 2012; September 11, 2012; October 9, 2012, February 25, 2013.

Judy Westrick, PhD, Tamarack Environmental Laboratories, LLC. September 24, 2012; December 17, 2012; January 10, 2013.

Tom McNabb, Clean Lakes, Inc. September 1, 2012; January 28, 2013.

APPENDIX A

Summary Tables of Sampling Locations and Data

This appendix contains summary tables for sampling location coordinates, sampling times, dissolved oxygen (DO) measurements, DO saturation, water temperature, turbidity, Secchi disk readings, reservoir depth, and field data measurements.

A.1 Sampling Location Coordinates

Each sampling location was identified using a Garmin Oregon® 450 Geographic Positioning System (GPS) prior to pre-treatment sampling (Table A-1). The coordinates were recorded in the GPS and later used to position the boat when subsequent samples were collected. This procedure ensured that the location of the pre-event, event, post-event, and one-week sampling would consistent. There was some minor variability in terms of sampling locations.

Table A-1. Average approximate coordinates of sampling locations.

Sampling Location	Coordinates	
N1	41°56’40.81”N	122°25’34.66”W
N2	41°56’37.86”N	122°25’33.85”W
N3	41°56’35.30”N	122°25’32.56”W
T1	41°56’40.25”N	122°25’27.77”W
T2	41°56’37.68”N	122°25’27.59”W
T3	41°56’39.14”N	122°25’25.61”W

A.2 Sampling Times

Sampling occurred at four times: the morning prior to application (“pre-event”), immediately after application (“event”), the following morning (“post-event”), and one week after application (“one-week”). Sampling times for each location and depth are summarized below.

Table A-2. Summary of sampling times, September event.

Location	Depth (m)	Sampling Date and Time			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	11:05	13:00	10:25	11:17
N1	Integrated	11:16	13:15	10:33	11:33
N2	0.1	11:30	13:25	11:00	12:07
N2	Integrated	11:44	13:33	11:05	12:16
N3	0.1	11:55	13:45	11:17	12:30
N3	Integrated	12:04	13:50	11:21	12:40
T1	0.1	09:10	14:05	11:44	13:15
T1	Integrated	09:25	14:15	11:50	13:25
T2	0.1	10:00	14:28	12:05	13:35
T2	Integrated	10:15	14:33	12:15	13:50
T3	0.1	10:35	14:45	12:22	14:07
T3	Integrated	10:47	14:56	12:30	14:15

Table A-3. Summary of sampling times, October event.

Location	Depth (m)	Sampling Date and Time			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-event (10/3/2013)	One-week (10/9/2013)
N1	0.1	11:48	15:36	11:44	11:35
N1	Integrated	11:57	15:30	11:46	11:45
N2	0.1	12:16	15:49	12:02	12:10
N2	Integrated	12:23	15:54	12:08	12:20
N3	0.1	12:45	16:07	12:29	12:35
N3	Integrated	12:51	16:13	12:31	12:45
T1	0.1	10:00	13:52	10:29	13:00
T1	Integrated	10:16	14:01	10:34	13:10
T2	0.1	10:41	14:20	10:47	13:25
T2	Integrated	10:47	14:27	10:55	13:30
T3	0.1	11:10	14:43	11:08	13:50
T3	Integrated	11:17	14:52	11:13	14:00

A.2 Temperature

There was no discernible change in water temperature related to the algaecide application. Rather, changes were in response to daily thermal dynamics of the reservoir and atmosphere. Water temperature readings were collected with an In-Situ, Inc., Troll 9500 Professional (#45654) water quality probe. The temperature/conductivity sensor has an operational range of -5°C to 75°C in water. The results have an accuracy of ±0.2°C.

Figure A-1. Water temperature measurements, September event.

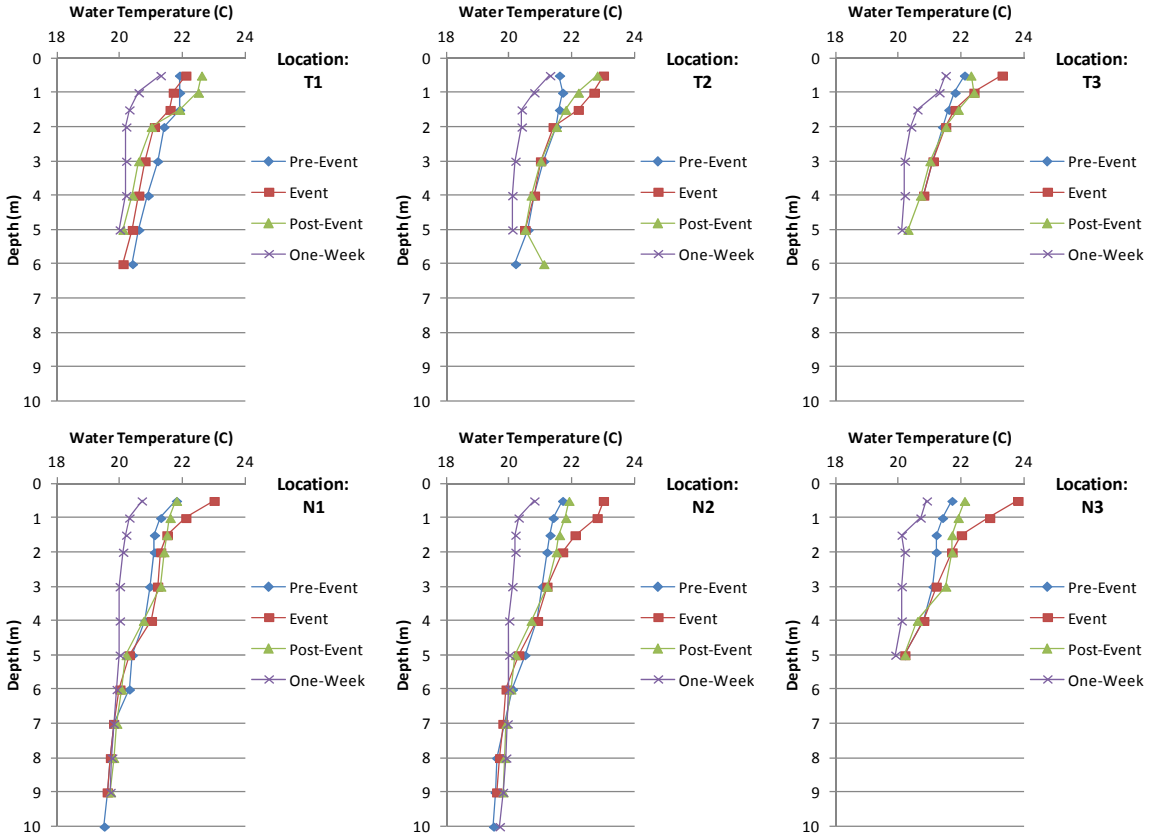
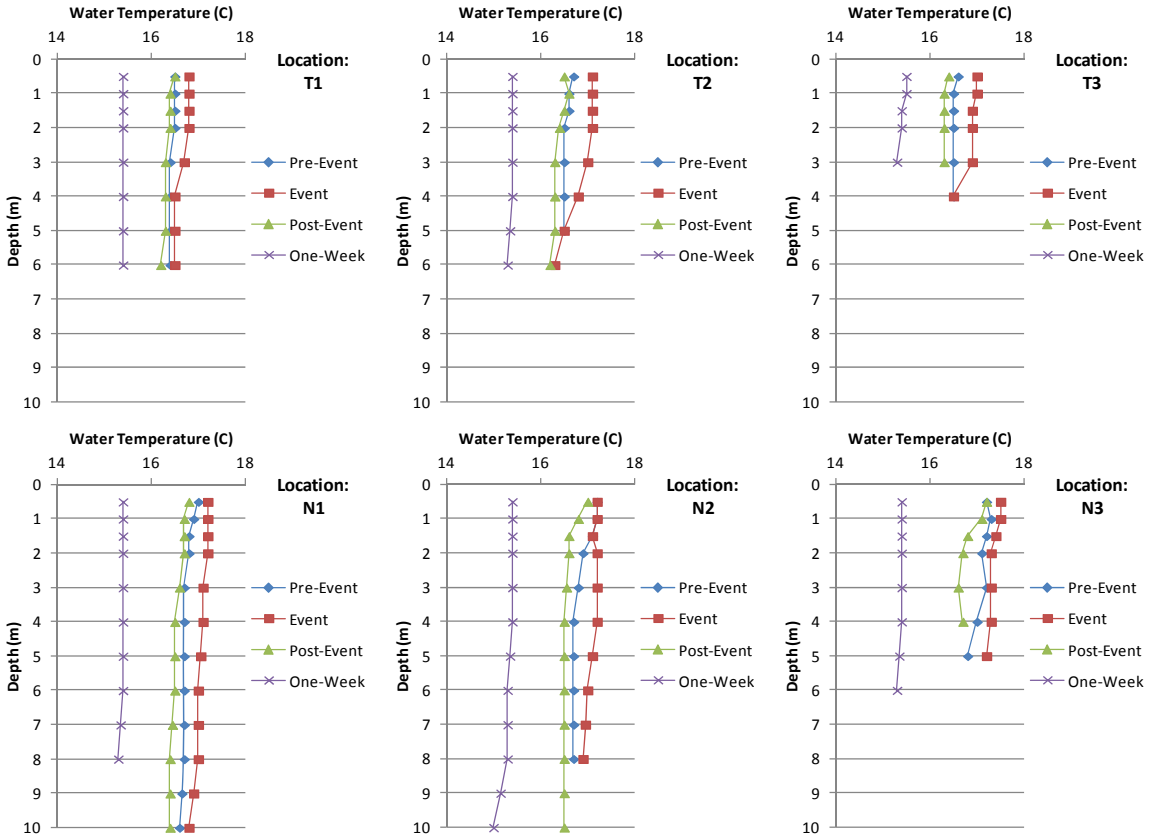


Figure A-2. Water temperature measurements, October event. (Note the scale change from the September event.)



A.3 Dissolved Oxygen (DO)

Dissolved oxygen (DO) levels ranged from super-saturation to sub-saturation depending on location and depth. As such, DO at certain locations at certain times was below water quality criteria (Figure A-3 through A-6). However, treatment activities did not further reduce DO levels. Rather, local conditions associated with primary production, stratification, meteorology, and wind and other mixing processes governed DO concentrations. A review of Figure A-1 through A-4 indicates that pre- and post-sampling profiles were largely consistent within the treatment area and in the control sites outside the treatment area. From the first treatment in September the last treatment in October the lake became isothermal in the study area with DO conditions within the treatment area higher than those outside the treatment area in the main lake. DO readings were collected with an In-Situ, Inc., Troll 9500 Professional (#45654) water quality probe. The DO measurements have an accuracy of ±2 percent of reading or ±0.2 mg/L, whichever is greater.

Figure A-3. Dissolved oxygen concentrations (mg/L), September event.

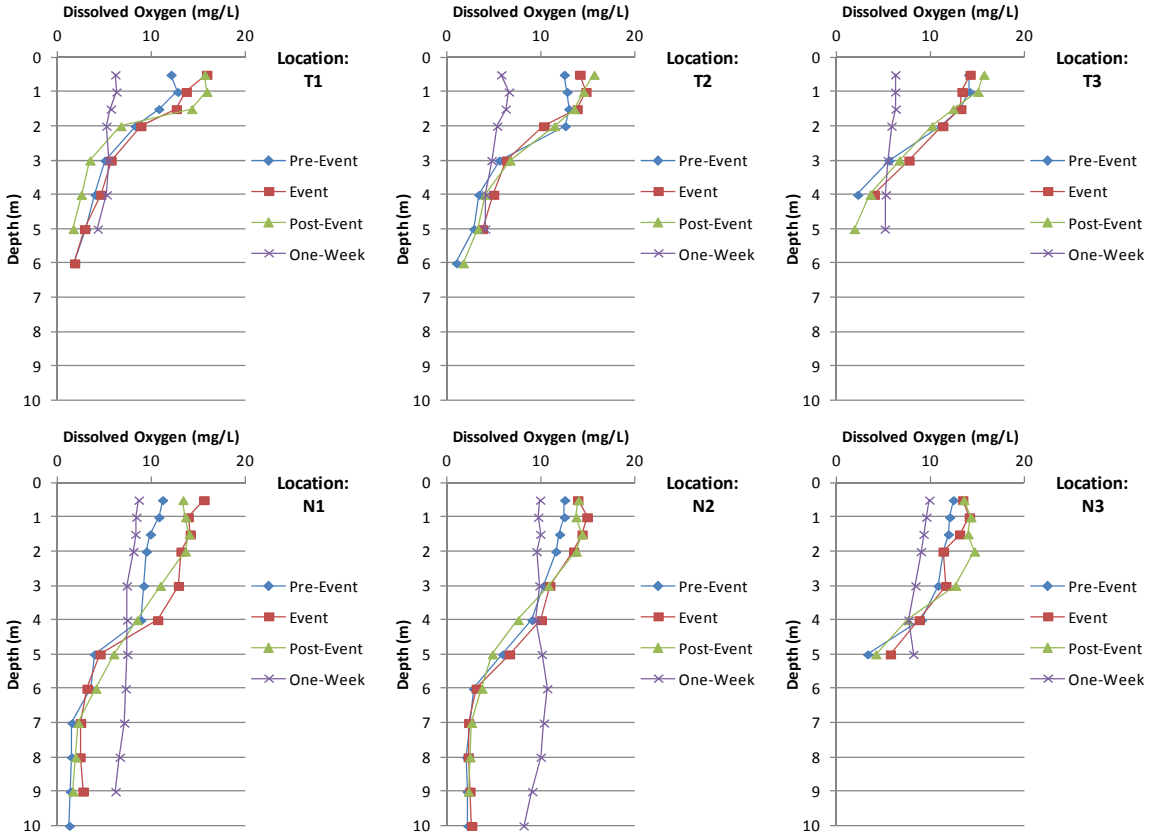


Figure A-4. Dissolved oxygen percent saturation (%), September event.

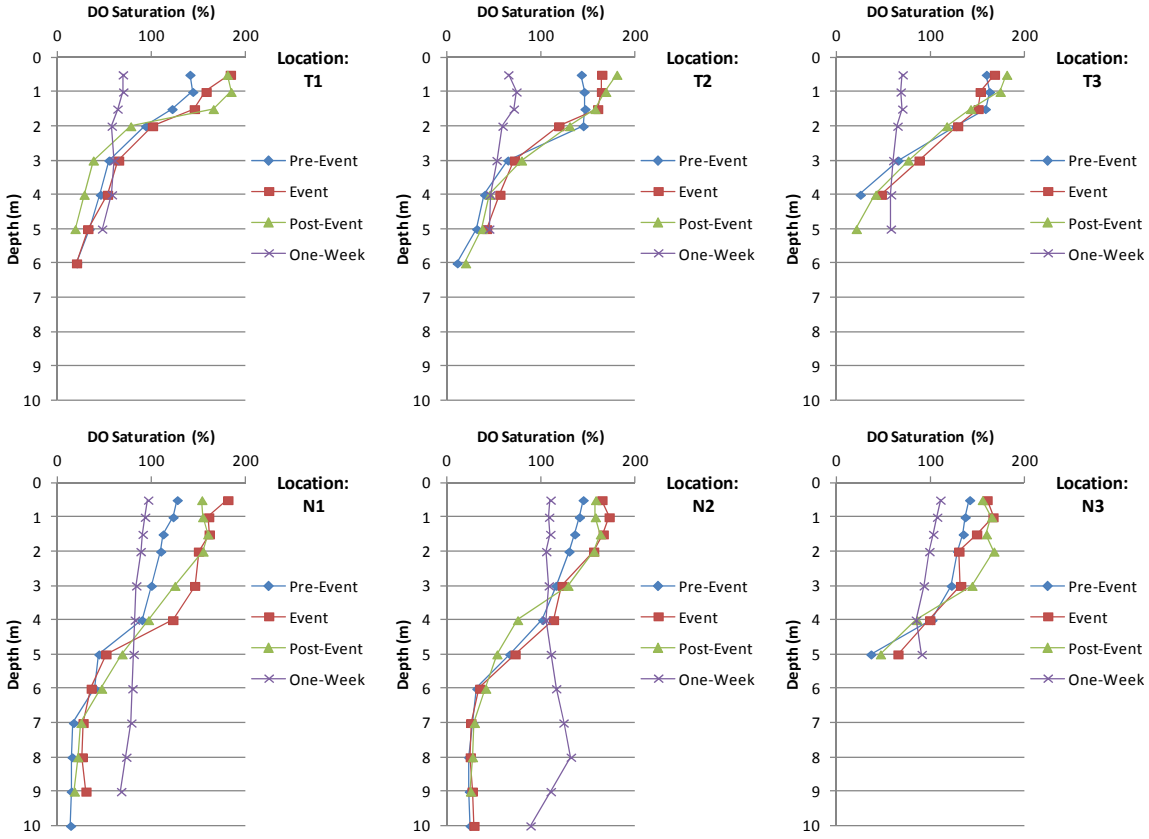


Figure A-5. Dissolved oxygen concentrations (mg/L), October event. (Note the scale change from the September event.)

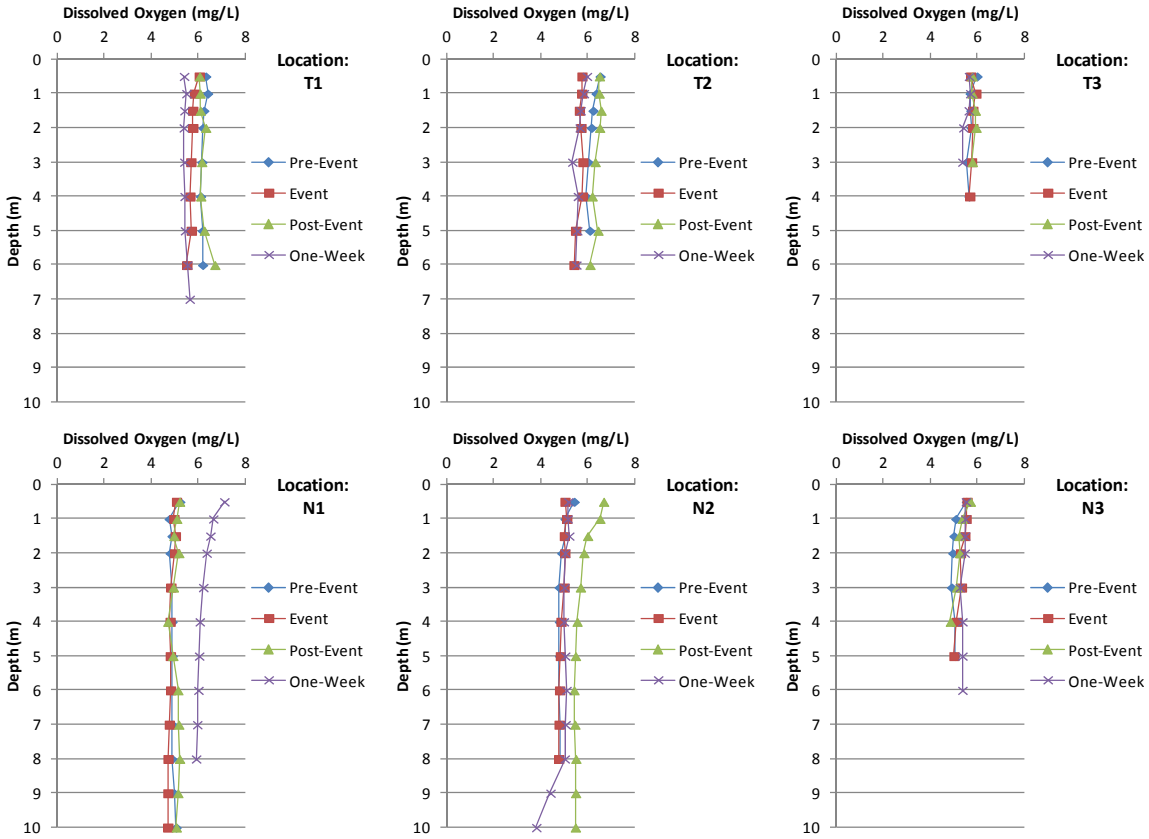
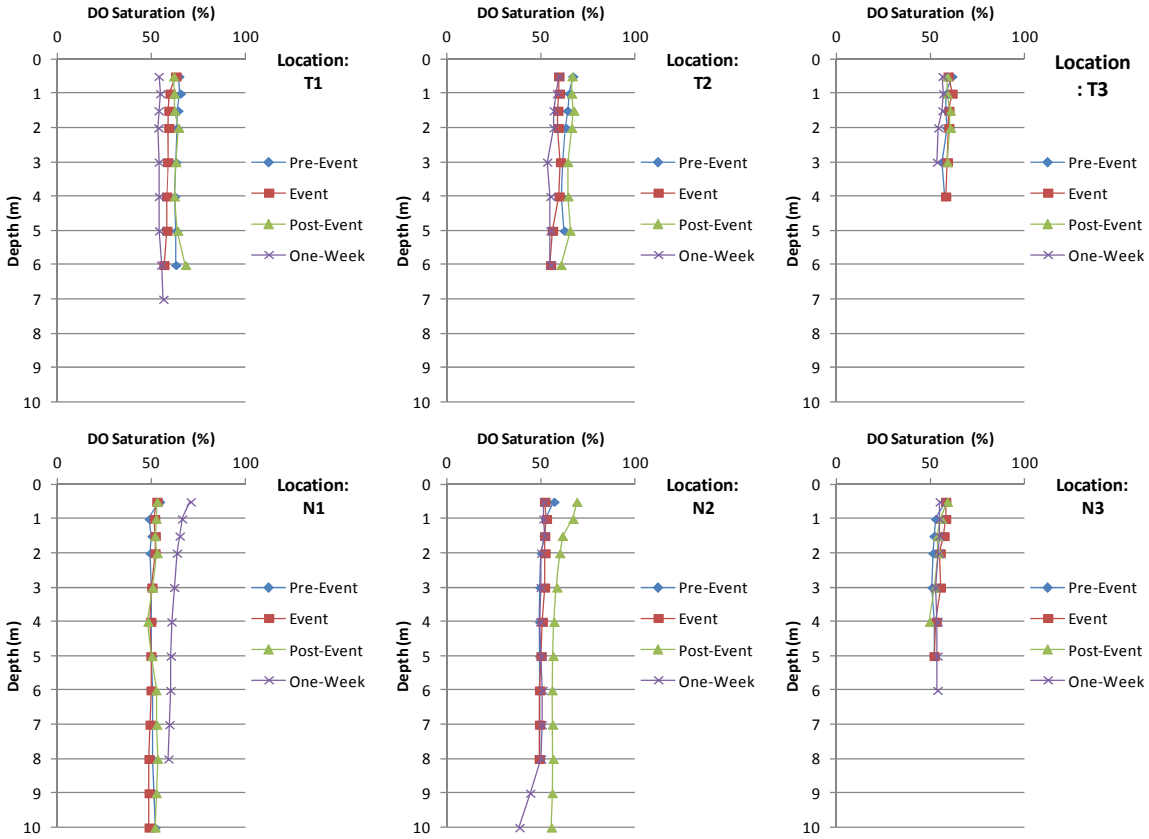


Figure A-6. Dissolved oxygen percent saturation (%), October event. (Note the scale change from the September event.)



A.4 pH

pH was above the water quality standard throughout the study, which is routinely above the 7.0 to 8.5 standard throughout much of the Klamath River system in summer periods and was in excess of 8.0 throughout the water column prior to algaecide application) (Figure A-7 and A-8). According to the manufacturer, application of GreenClean Liquid is not expected to considerably change pH (pers. comm. V. Choppakatla, February 25, 2013). This has been confirmed through a review of expected pH changes as determined from BioSafe bench top studies (BioSafe 2009) on GreenClean Liquid, wherein distilled water, pond water, and groundwater were tested with GreenClean Liquid to determine pH response. Further, a review of pH measurements in previous PacifiCorp algaecide studies using GreenClean Liquid indicates that pH in discrete containers exhibited changes in the range of 0.03 to 0.04 pH units after algaecide application (Watercourse 2012). Based on discussions with BioSafe, review of prior BioSafe and PacifiCorp algaecide study data, and considering the dilution of the GreenClean product within the reservoir as well as the buffering capacity of natural waters (due to alkalinity and organic matter), significant pH changes were not expected, nor were they observed in the event and post-event data. pH readings were collected with an In-Situ, Inc., Troll 9500 Professional (#45654) water quality probe. The pH sensor has an accuracy of ±0.2.

Figure A-7. pH measurements, September event.

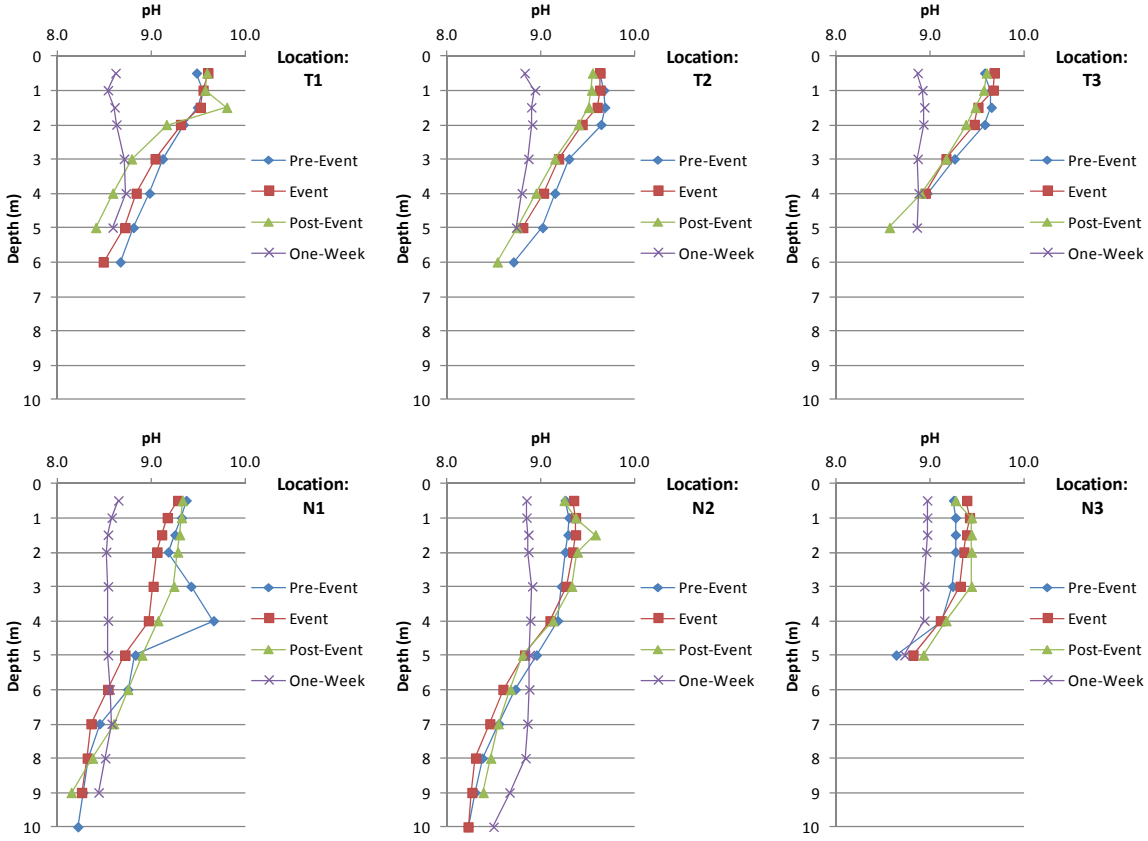
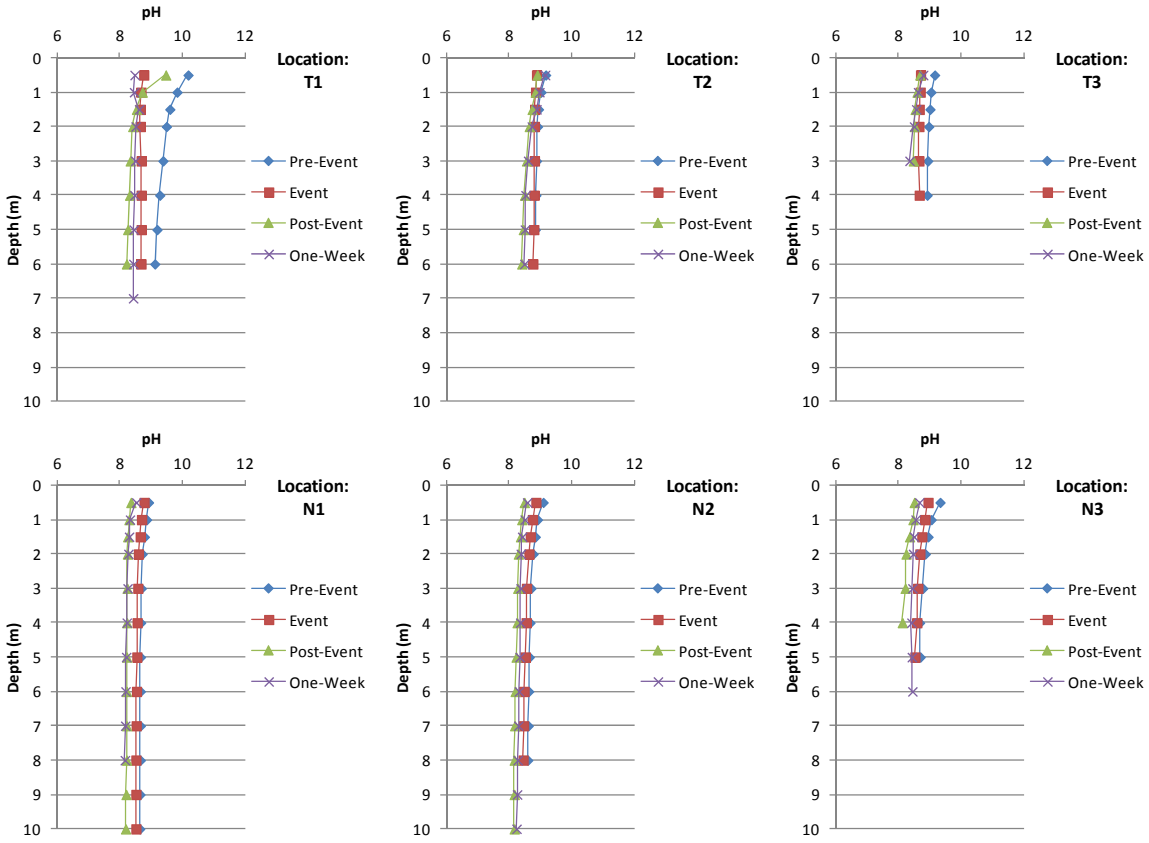


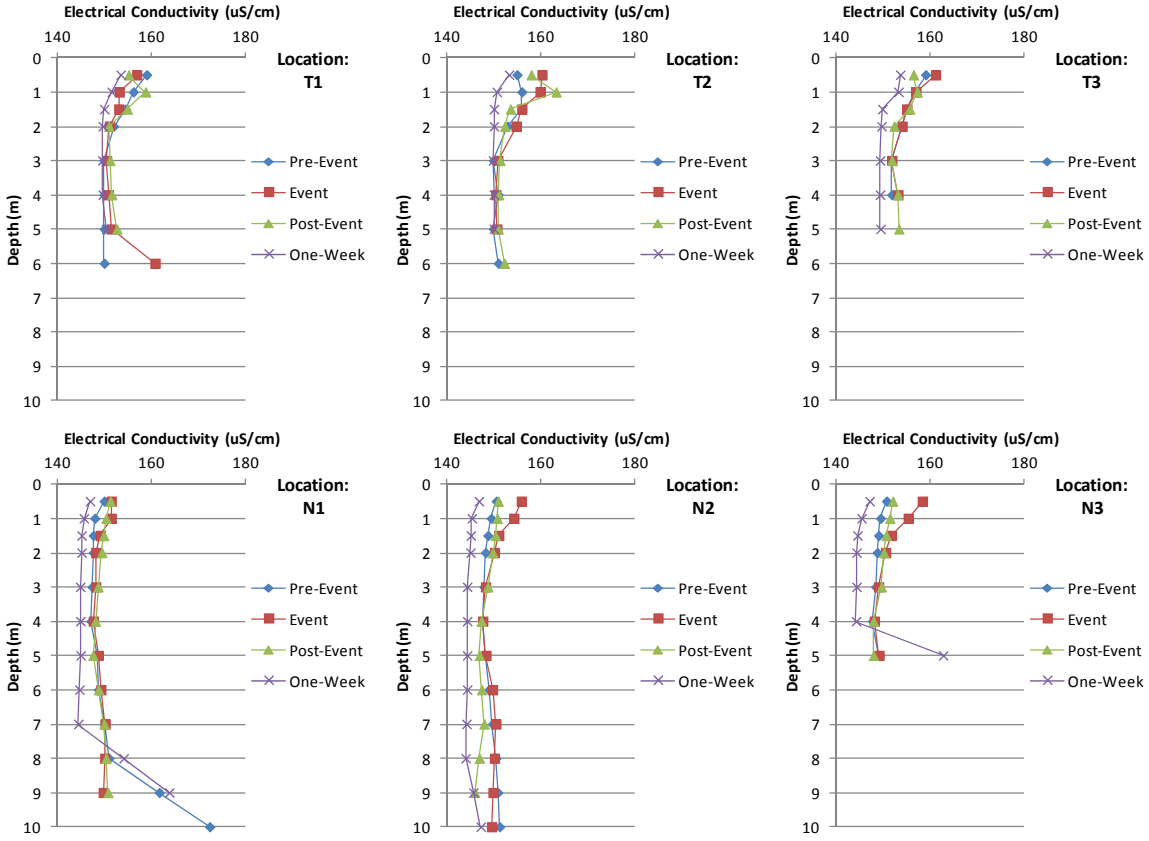
Figure A-8. pH measurements, October event. (Note the scale change from the September event.)



A.5 Electrical Conductivity

Conductivity remained stable throughout the pilot study (Figure A-9). Electrical conductivity readings were collected with an In-Situ, Inc., Troll 9500 Professional (#45654) water quality probe. The temperature/conductivity sensor has an accuracy of ±1 µS or ±1% of reading, whichever is greater. Electrical conductivity readings were only collected during the September event.

Figure A-9. Electrical conductivity measurements, September event.



A.6 Turbidity

During the September event, turbidity was less in the integrated samples for both the control and application sites (Table A-4). By the October event, turbidity was lower in control sites than the application sites (Table A-5).

Table A-4. Summary of turbidity measurements, September event.

Location	Depth (m)	Turbidity (NTU) Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.5	12.5	54.3	17.4	3.0
N1	Integrated	4.9	4.7	11.1	4.5
N2	0.5	7.0	33.7	14.5	10.2
N2	Integrated	8.4	9.6	6.1	8.4
N3	0.5	24.0	16.0	21.0	25.3
N3	Integrated	13.5	7.5	5.3	8.8
T1	0.5	39.5	24.9	14.6	4.1
T1	Integrated	13.4	13.5	12.2	6.5
T2	0.5	27.7	21.5	16.7	9.0
T2	Integrated	14.6	15.9	11.3	4.8
T3	0.5	42.5	24.2	19.3	15.3
T3	Integrated	26.5	21.4	12.5	11.0

*The Hach® 2100Q Portable Turbidimeter has an accuracy of ±2% of reading.

Table A-5. Summary of turbidity measurements, October event.

Location	Depth (m)	Turbidity (NTU) Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.5	1.5	3.0	1.8	2.1
N1	Integrated	2.1	1.9	1.9	2.3
N2	0.5	1.9	1.7	1.7	1.9
N2	Integrated	2.6	2.0	3.3	2.1
N3	0.5	2.9	2.6	3.6	2.0
N3	Integrated	6.2	2.4	3.1	2.1
T1	0.5	9.1	6.4	8.1	3.4
T1	Integrated	4.8	11.4	8.3	2.0
T2	0.5	7.4	14.4	11.6	2.5
T2	Integrated	6.5	12.5	6.8	2.0
T3	0.5	10.1	33.9	7.4	2.0
T3	Integrated	9.1	8.2	7.7	1.9

*The Hach® 2100Q Portable Turbidimeter has an accuracy of ±2% of reading.

A.7 Depths

Secchi depths were taken at most sampling locations and times.

Table A-6. Summary of Secchi depth measurements, September event.

	Secchi Depth (m)			
Location	**Pre-Event (9/11/2013)**	**Event (9/11/2013)**	**Post-Event (9/12/2013)**	**One-Week (9/18/2013)**
N1	1.75	0.75	1.25	2.00
N2	1.25	1.00	1.00	1.75
N3	1.25	1.00	0.75	0.75
T1	-	1.00	1.25	1.75
T2	1.00	0.75	0.75	1.75
T3	0.75	0.75	1.00	1.50

Table A-7. Summary of Secchi depth measurements, October event.

	Secchi Depth (m)			
Location	**Pre-Event (10/2/2013)**	**Event (10/2/2013)**	**Post-Event (10/3/2013)**	**One-Week (10/9/2013)**
N1	3.00	2.75	3.00	2.75
N2	2.00	2.50	3.25	2.75
N3	3.25	2.25	2.25	2.50
T1	2.00	1.75	1.75	2.50
T2	1.75	1.75	1.50	3.00
T3	2.00	1.25	2.00	2.50

A.9 Field Data

This section summarizes the field data that was collected and analyzed for the 2013 algaecide study.

Table A-8. Summary of total Nitrogen (TN) measurements, September event.

		Total Nitrogen (mg/L)			
Location	**Depth (m)**	**Pre-Event (9/11/2013)**	**Event (9/11/2013)**	**Post-event (9/12/2013)**	**One-week (9/18/2013)**
N1	0.1	1.37	2.73	1.44	1.48
N1	Integrated	1.56	1.47	1.48	1.47
N2	0.1	1.21	1.62	1.47	2.02
N2	Integrated	1.41	1.46	1.75	1.33
N3	0.1	1.25	1.86	1.88	2.55
N3	Integrated	1.25	1.30	1.47	1.25
T1	0.1	1.79	1.79	1.45	1.28
T1	Integrated	1.48	1.61	1.37	1.36
T2	0.1	1.80	1.56	1.65	1.42
T2	Integrated	1.41	1.37	1.38	1.30
T3	0.1	1.95	1.85	1.54	1.33
T3	Integrated	1.46	1.70	1.47	1.34

Table A-9. Summary of total Nitrogen (TN) measurements, October event.

		Total Nitrogen (mg/L)			
Location	**Depth (m)**	**Pre-Event (10/2/2013)**	**Event (10/2/2013)**	**Post-Event (10/3/2013)**	**One-Week (10/9/2013)**
N1	0.1	0.99	0.96	1.41	0.97
N1	Integrated	1.27	0.97	0.99	1.22
N2	0.1	1.53	0.93	0.99	1.02
N2	Integrated	1.20	3.89	1.00	1.02
N3	0.1	0.97	0.96	1.11	1.08
N3	Integrated	2.61	2.79	1.10	1.40
T1	0.1	1.45	1.12	1.38	1.16
T1	Integrated	1.44	1.26	1.63	1.08
T2	0.1	1.56	1.79	0.93	1.03
T2	Integrated	1.38	2.01	1.38	0.95
T3	0.1	1.63	0.97	1.64	1.09
T3	Integrated	1.47	1.45	1.71	0.92

Table A-10. Summary of Nitrate+Nitrite (NO2+NO3) measurements, September event.

Location	Depth (m)	Total Nitrate+Nitrite (mg/L)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	0.18	0.14	0.16	0.16
N1	Integrated	0.18	0.14	0.16	0.12
N2	0.1	0.19	0.15	0.14	0.13
N2	Integrated	0.18	0.13	0.14	0.08
N3	0.1	0.19	0.13	0.16	0.10
N3	Integrated	0.18	0.13	0.15	0.05
T1	0.1	0.13	0.18	0.18	0.04
T1	Integrated	0.13	0.18	0.18	0.04
T2	0.1	0.13	0.19	0.19	0.04
T2	Integrated	0.13	0.19	0.18	0.04
T3	0.1	0.13	0.18	0.18	0.16
T3	Integrated	0.13	0.18	0.18	0.03

Table A-11. Summary of Nitrate+Nitrite (NO2+NO3) measurements, October event.

Location	Depth (m)	Total Nitrate+Nitrite (mg/L)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	0.17	0.18	0.17	0.20
N1	Integrated	0.17	0.18	0.17	0.20
N2	0.1	0.18	0.18	0.17	0.20
N2	Integrated	0.18	0.18	0.17	0.20
N3	0.1	0.18	0.17	0.17	0.20
N3	Integrated	0.18	0.17	0.17	0.20
T1	0.1	0.12	0.12	0.15	0.16
T1	Integrated	0.12	0.12	0.14	0.16
T2	0.1	0.12	0.14	0.13	0.16
T2	Integrated	0.12	0.13	0.13	0.16
T3	0.1	0.12	0.11	0.15	0.16
T3	Integrated	0.12	0.12	0.14	0.16

Table A-12. Summary of Ammonia (NH4) measurements, September event.

Location	Depth (m)	Total Ammonia (mg/L)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	0.23	0.22	0.22	0.03
N1	Integrated	0.23	0.22	0.22	0.05
N2	0.1	0.22	0.20	0.21	0.01
N2	Integrated	0.22	0.20	0.22	0.03
N3	0.1	0.22	0.20	0.22	0.01
N3	Integrated	0.22	0.21	0.22	0.03
T1	0.1	0.22	0.22	0.25	0.13
T1	Integrated	0.23	0.22	0.23	0.14
T2	0.1	0.17	0.21	0.22	0.12
T2	Integrated	0.19	0.20	0.23	0.14
T3	0.1	0.22	0.20	0.21	0.11
T3	Integrated	0.23	0.20	0.20	0.10

Table A-13. Summary of Ammonia (NH4) measurements, October event.

Location	Depth (m)	Total Ammonia (mg/L)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	0.24	0.22	0.23	0.20
N1	Integrated	0.24	0.24	0.23	0.20
N2	0.1	0.22	0.20	0.24	0.19
N2	Integrated	0.23	0.22	0.23	0.20
N3	0.1	0.24	0.20	0.21	0.20
N3	Integrated	0.22	0.18	0.20	0.19
T1	0.1	0.22	0.22	0.22	0.24
T1	Integrated	0.24	0.23	0.21	0.25
T2	0.1	0.18	0.23	0.22	0.22
T2	Integrated	0.20	0.21	0.24	0.23
T3	0.1	0.22	0.20	0.22	0.22
T3	Integrated	0.22	0.20	0.23	0.23

Table A-14. Summary of total Phosphorus (TP) measurements, September event.

Location	Depth (m)	Total Phosphorus (mg/L)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	0.16	0.30	0.19	0.17
N1	Integrated	0.19	0.17	0.16	0.17
N2	0.1	0.15	0.23	0.18	0.22
N2	Integrated	0.17	0.16	0.19	0.17
N3	0.1	0.18	0.22	0.35	0.29
N3	Integrated	0.15	0.16	0.17	0.15
T1	0.1	0.17	0.24	0.23	0.20
T1	Integrated	0.21	0.20	0.22	0.20
T2	0.1	0.18	0.24	0.28	0.20
T2	Integrated	0.16	0.23	0.22	0.21
T3	0.1	0.24	0.25	0.23	0.21
T3	Integrated	0.21	0.23	0.21	0.19

Table A-15. Summary of total Phosphorus (TP) measurements, October event.

Location	Depth (m)	Total Phosphorus (mg/L)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	0.15	0.15	0.27	0.16
N1	Integrated	0.13	0.18	0.17	0.20
N2	0.1	0.22	0.17	0.18	0.13
N2	Integrated	0.29	0.79	0.17	0.18
N3	0.1	0.15	0.15	0.21	0.15
N3	Integrated	0.53	0.52	0.18	0.25
T1	0.1	0.17	0.19	0.25	0.19
T1	Integrated	0.16	0.25	0.30	0.18
T2	0.1	0.25	0.31	0.16	0.17
T2	Integrated	0.15	0.29	0.23	0.14
T3	0.1	0.19	0.19	0.27	0.16
T3	Integrated	0.28	0.26	0.24	0.13

Table A-16. Summary of Phosphate (PO4) measurements, September event.

Location	Depth (m)	Total Phosphate (mg/L)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	0.09	0.09	0.09	0.12
N1	Integrated	0.11	0.11	0.11	0.11
N2	0.1	0.09	0.09	0.09	0.11
N2	Integrated	0.11	0.11	0.11	0.11
N3	0.1	0.09	0.09	0.10	0.10
N3	Integrated	0.10	0.10	0.09	0.11
T1	0.1	0.07	0.08	0.09	0.14
T1	Integrated	0.10	0.11	0.13	0.13
T2	0.1	0.07	0.08	0.09	0.13
T2	Integrated	0.11	0.11	0.13	0.13
T3	0.1	0.06	0.08	0.09	0.13
T3	Integrated	0.09	0.11	0.11	0.12

Table A-17. Summary of Phosphate (PO4) measurements, October event.

Location	Depth (m)	Total Phosphate (mg/L)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	0.13	0.13	0.13	0.13
N1	Integrated	0.13	0.13	0.14	0.13
N2	0.1	0.13	0.13	0.13	0.13
N2	Integrated	0.13	0.13	0.13	0.13
N3	0.1	0.13	0.13	0.13	0.13
N3	Integrated	0.13	0.13	0.13	0.13
T1	0.1	0.13	0.13	0.13	0.13
T1	Integrated	0.12	0.13	0.13	0.13
T2	0.1	0.12	0.12	0.13	0.13
T2	Integrated	0.12	0.12	0.12	0.13
T3	0.1	0.13	0.12	0.13	0.13
T3	Integrated	0.12	0.13	0.13	0.13

Table A-18. Summary of dissolved organic carbon (DOC) measurements, September event.

Location	Depth (m)	Total Dissolved Organic Carbon (ppm)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	6.41	6.24	6.62	6.46
N1	Integrated	7.14	7.15	7.47	7.60
N2	0.1	6.20	6.38	6.39	6.21
N2	Integrated	7.05	6.70	7.06	6.98
N3	0.1	6.16	6.20	6.95	6.29
N3	Integrated	6.74	6.86	6.57	7.02
T1	0.1	6.99	8.40	6.99	6.66
T1	Integrated	7.64	7.61	7.23	7.35
T2	0.1	6.61	8.20	7.23	7.37
T2	Integrated	6.99	8.34	7.26	7.37
T3	0.1	6.78	8.26	7.04	7.46
T3	Integrated	6.89	7.63	7.22	6.64

Table A-19. Summary of dissolved organic carbon (DOC) measurements, October event.

Location	Depth (m)	Total Dissolved Organic Carbon (ppm)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	5.52	5.76	5.49	5.53
N1	Integrated	5.95	5.90	5.90	5.93
N2	0.1	5.57	5.43	5.55	5.57
N2	Integrated	6.25	5.78	5.72	6.09
N3	0.1	5.52	5.51	5.58	5.62
N3	Integrated	5.85	5.71	5.84	5.98
T1	0.1	5.77	5.84	5.63	5.70
T1	Integrated	6.10	6.18	6.28	5.83
T2	0.1	5.48	5.80	5.63	5.58
T2	Integrated	5.97	5.85	6.24	5.95
T3	0.1	5.57	5.80	5.87	5.70
T3	Integrated	6.07	6.03	5.93	5.84

Table A-20. Summary of Chlorophyll *a* measurements, September event.

Location	Depth (m)	Total Chlorophyll *a* (ppb)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	33.4	145.2	43.6	7.3
N1	Integrated	19.6	29.0	15.3	5.9
N2	0.1	26.9	36.3	45.8	18.4
N2	Integrated	33.4	28.3	22.5	9.2
N3	0.1	36.0	45.8	45.8	26.8
N3	Integrated	31.2	21.8	36.3	10.9
T1	0.1	55.2	38.5	14.5	2.5
T1	Integrated	24.0	27.6	10.3	3.6
T2	0.1	56.6	38.5	11.4	7.5
T2	Integrated	30.5	24.0	10.9	6.1
T3	0.1	74.1	39.2	9.5	7.0
T3	Integrated	36.0	28.3	4.2	6.1

Table A-21. Summary of Chlorophyll *a* measurements, October event.

Location	Depth (m)	Total Cholorphyll *a* (ppb)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	4.1	3.3	4.7	1.9
N1	Integrated	2.7	3.9	2.6	2.4
N2	0.1	8.9	3.9	3.6	3.0
N2	Integrated	3.3	5.4	1.4	3.1
N3	0.1	3.6	10.0	7.0	3.5
N3	Integrated	3.9	5.0	8.4	3.7
T1	0.1	5.9	16.7	32.0	2.8
T1	Integrated	5.3	14.5	29.0	3.8
T2	0.1	2.5	31.2	39.2	8.9
T2	Integrated	7.5	27.6	18.9	8.6
T3	0.1	7.8	168.5	20.3	7.3
T3	Integrated	5.3	34.1	29.0	5.6

Table A-22. Summary of Microcystin measurements, September event.

Location	Depth (m)	Total Microcystin (ug/L)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	23.1	78.6	26.9	15.1
N1	Integrated	118.0	177.6	8.0	15.3
N2	0.1	21.5	38.2	36.0	36.9
N2	Integrated	26.0	22.5	18.6	20.0
N3	0.1	24.3	39.3	19.8	98.1
N3	Integrated	202.2	21.0	38.1	23.7
T1	0.1	52.5	10.2	21.4	17.4
T1	Integrated	102.2	20.4	267.2	13.6
T2	0.1	45.8	23.1	25.3	15.9
T2	Integrated	22.9	21.3	116.2	17.6
T3	0.1	32.3	21.6	19.5	4.1
T3	Integrated	17.0	11.7	11.9	23.7

Table A-23. Summary of Microcystin measurements, October event.

Location	Depth (m)	Total Microcystin (ug/L)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	2.5	2.8	2.5	0.3
N1	Integrated	2.2	2.5	3.4	1.1
N2	0.1	4.1	1.9	2.2	1.5
N2	Integrated	2.2	4.1	1.8	1.6
N3	0.1	2.1	4.4	3.2	0.7
N3	Integrated	1.7	3.2	4.2	2.3
T1	0.1	8.6	4.3	10.9	2.0
T1	Integrated	9.7	5.2	20.6	2.9
T2	0.1	18.1	16.3	25.7	3.8
T2	Integrated	22.8	15.9	15.3	3.9
T3	0.1	25.9	88.5	19.6	4.0
T3	Integrated	14.0	17.5	21.5	3.3

Table A-24. Summary of *Microcystis aeruginosa* (MSAE) measurements, September event.

Location	Depth (m)	Total MSAE (1,000 cells/mL)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	155	733	441	73
N1	Integrated	37	86	42	45
N2	0.1	102	186	248	364
N2	Integrated	177	80	69	71
N3	0.1	188	211	149	413
N3	Integrated	113	75	429	70
T1	0.1	352	299	113	42
T1	Integrated	114	130	89	41
T2	0.1	486	338	104	59
T2	Integrated	174	118	127	47
T3	0.1	465	265	193	41
T3	Integrated	148	197	99	59

Table A-25. Summary of *Microcystis aeruginosa* (MSAE) measurements, October event.

Location	Depth (m)	Total MSAE (1,000 cells/mL)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	3	28	9	2
N1	Integrated	10	10	7	1
N2	0.1	21	5	-	1
N2	Integrated	4	8	7	1
N3	0.1	18	12	22	2
N3	Integrated	8	22	35	1
T1	0.1	68	38	105	8
T1	Integrated	66	46	77	11
T2	0.1	120	182	133	24
T2	Integrated	69	59	49	21
T3	0.1	79	751	68	14
T3	Integrated	75	111	83	8

Table A-26. Summary of *Aphanizomenon flos-aquae* (APHA) measurements, September event.

Location	Depth (m)	Total APHA (cells/mL)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	20634	12674	8395	-
N1	Integrated	24401	14718	14843	416
N2	0.1	24805	5154	14476	1513
N2	Integrated	55200	25118	6653	16616
N3	0.1	34451	13132	9155	5089
N3	Integrated	14684	39391	12035	8095
T1	0.1	8109	5324	6367	-
T1	Integrated	8054	7543	1322	3808
T2	0.1	8668	9716	5026	-
T2	Integrated	3218	5246	2765	755
T3	0.1	7530	9730	9187	-
T3	Integrated	5678	4867	1637	-

Table A-27. Summary of *Aphanizomenon flos-aquae* (APHA) measurements, October event.

Location	Depth (m)	Total APHA (cells/mL)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	-	-	-	-
N1	Integrated	-	-	-	-
N2	0.1	621	-	-	-
N2	Integrated	-	1634	-	161
N3	0.1	-	-	-	-
N3	Integrated	-	-	6560	-
T1	0.1	292	1715	705	-
T1	Integrated	304	150	-	1658
T2	0.1	1381	-	-	-
T2	Integrated	-	634	399	-
T3	0.1	-	477	2946	-
T3	Integrated	-	1230	617	-

Table A-28. Summary of *Anabaena sp.* (ABXX) measurements, September event.

Location	Depth (m)	Total ABXX (cells/mL)			
		Pre-Event (9/11/2013)	Event (9/11/2013)	Post-event (9/12/2013)	One-week (9/18/2013)
N1	0.1	1368	1102	988	-
N1	Integrated	1851	1554	370	-
N2	0.1	-	-	505	504
N2	Integrated	-	2952	-	565
N3	0.1	940	-	1010	-
N3	Integrated	1114	1142	912	-
T1	0.1	-	-	-	-
T1	Integrated	-	349	-	498
T2	0.1	-	-	-	-
T2	Integrated	216	-	-	-
T3	0.1	-	-	-	-
T3	Integrated	-	-	-	426

Table A-29. Summary of *Anabaena sp.* (ABXX) measurements, October event.

Location	Depth (m)	Total ABXX (cells/mL)			
		Pre-Event (10/2/2013)	Event (10/2/2013)	Post-Event (10/3/2013)	One-Week (10/9/2013)
N1	0.1	-	-	-	-
N1	Integrated	-	-	-	-
N2	0.1	-	-	-	-
N2	Integrated	-	-	-	-
N3	0.1	-	-	-	-
N3	Integrated	-	-	-	-
T1	0.1	-	-	-	-
T1	Integrated	-	-	-	-
T2	0.1	307	-	-	-
T2	Integrated	-	-	-	-
T3	0.1	-	-	-	-
T3	Integrated	-	-	-	-