Assessment of Lyngbya Treatment Program: Determining Potential for Early Season Control, Impacts of Continual Annual Treatments, and Algaecide Treatment Efficacy

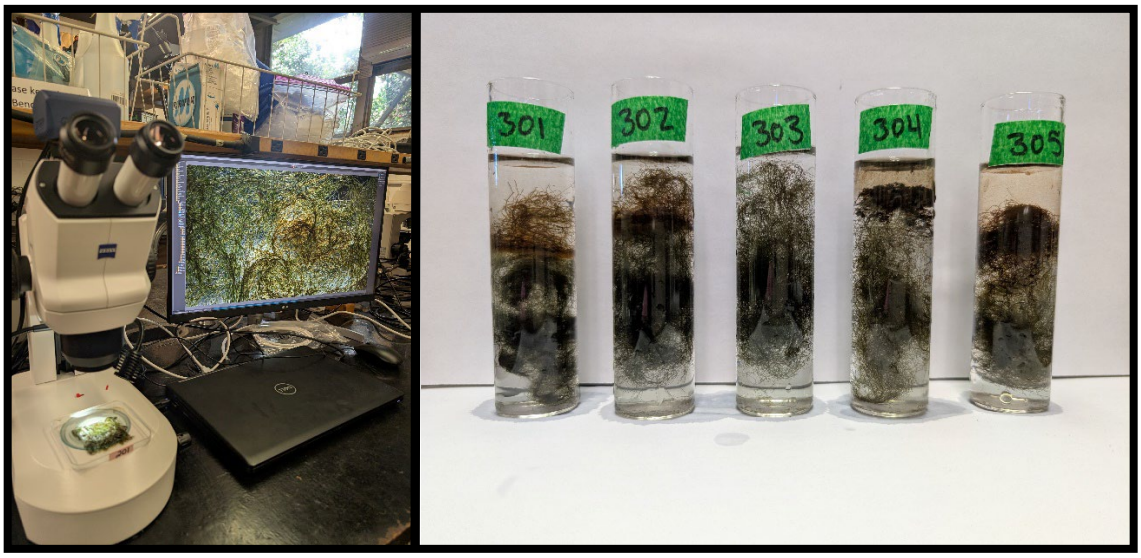
2025

Prepared By:

North Carolina State University
Aquatic Plant Management Program
Raleigh, NC 27607

Submitted To:

Lake Gaston Weed Control Council
Littleton, NC 27850



NC STATE UNIVERSITY

Introduction

The efficacy of algaecides for controlling filamentous algae have been well described (Bishop et al. 2015; Calomeni et al. 2018; Rogers and Pietruszewski 2021). However, there are many site-specific challenges that can influence the efficacy of algaecide treatments. Factors such as algae morphology and phenology, as well as ecological factors like water temperature and quality can greatly effect algaecide efficacy (Lembi 2000). Additionally, the development of herbicide resistance presents a growing concern in both agricultural systems and the management of vascular aquatic plants (Peterson 2017; Richardson 2008). For managers of lyngbya treatment programs there is also a need to balance the efficacy of their programs with potential negative impacts that chelated copper-based algaecides present to non-target aquatic organisms. To date, the potential of early season lyngbya treatments and the risk of decreased efficacy from repeated algaecide treatments has not been explored for lyngbya.

The objectives for the continued evaluation of chelated copper algaecides in a laboratory setting were to: 1) continue exploring the efficacy of varying algaecide combinations, 2) determine if seasonality and water temperature effects the efficacy of lyngbya treatments to allow for early season control opportunity, and 3) determine if multiple within-year algaecide treatments impacts the efficacy of consecutive yearly lyngbya treatments over time. These objectives were achieved through a series of three individual trials.

Methods

Standardized Laboratory Methods and Evaluations

For each trial, lyngbya collection, material preparation, and treatment followed the standard protocols outlined below. Table 1 provides an overview of the experimental set-ups for each of the three trials used to assess aspects of lyngbya treatment programs.

Lyngbya was collected from Lake Gaston and was brought back to NC State labs to evaluate initial vigor. Observable algal material displaying signs of decreased health or cell death was then removed and 0.25 g fresh weight of healthy lyngbya was placed in 40 mL test tubes containing 30 mL of Lake Gaston site water. Prior to treatment, vials of lyngbya were allowed to acclimate for approximately 5 days in ambient greenhouse conditions. During all acclimation and treatment time periods, the water temperature of lyngbya mesocosms was controlled by a water bath to simulate lake conditions at time of field collection.

All treatment units included an untreated control group and were represented by four replicates. Additional vials containing untreated lyngbya were evaluated prior to the beginning of some experiments to establish a baseline of health and were referred to as a pre-treatment harvest (PH). Treatment units and exposure times varied depending on the objective of the individual trial (Table 1). At the completion of the herbicide exposure period, lyngbya was gently removed from each vial, dabbed dry, suspended and washed in a beaker of fresh deionized water, removed, and patted dry again prior to being placed in sterile 40 mL test tube

containing 30 mL of untreated site water. Injury evaluations were performed 10 to 11-days post-treatment (depending on trial) (Table 1) to assess lyngbya response to each treatment combination.

To assess overall health, all lyngbya filaments within a given evaluation unit were spread evenly over a 12x12 cm area and visually evaluated at 40x magnification. Viability was determined by calculating the percent of green, healthy filaments versus those sheaths lacking viable cells or containing predominately chlorotic cells found in a single scope view. An average of six independent scope views was used to determine the overall percent viability for each unit. Visual assessment of each unit was performed by two independent readers and reassessment of viability was performed if differences in viability estimates were greater than 10 percent between the two readers. However, final viability levels were calculated from a single readers estimates to avoid pseudo replication.

To identify potential differences in treatment efficacy, an ANOVA was conducted. When treatment differences were identified, either Tukey's HSD test or Student's t-test were performed.

Algaecide Treatment Efficacy

Lyngbya samples were collected in June 2024 from Lake Gaston within an area outside of the current lyngbya treatment program. Treatment and evaluation protocols followed the methods previously described and treatment combinations included combinations of Captain XTR® + AMP, Cutrine Ultra® + AMP®, as well as Captain XTR® and Cutrine Ultra® applied independently. Treatments were applied at maximum label rates, with an exposure time of 72 hours. For this trial, evaluation of viability was performed 11-days post-treatment to assess lyngbya response to treatment and subsequent control. A series of three consecutive treatments occurred for each experimental unit and occurred at 3-week intervals.

Potential of Early Season Control

Independent lyngbya collections occurred at Lake Gaston during the months of February, March, April, and May within an area outside of the current lyngbya treatment program. Treatment protocols followed the methods previously described and treatment combinations included Captain XTR® + AMP, Cutrine Ultra® + AMP, as well as Captain XTR® and Cutrine Ultra® applied independently. Treatments were applied at maximum label rates, with an exposure time of 48 hours. Water bath conditions were held at 55°F in February, 60°F in March, 70°F in April, and 75°F in May.

Impacts of Continual Annual Treatments

To evaluate the impacts of continual annual algaecide treatments, lyngbya was collected from individual treatment sites in which initiation into the program occurred in either 2021, 2022, or 2024 and continual annual treatments have occurred. Due to a limited amount of treatment

acreage added in 2023, lyngbya samples were not robust enough for evaluation could not be obtained from these areas. All lyngbya samples were collected in June and therefore received two in-lake treatment applications as part of the 2024 season prior to collection. Collected samples represented areas of lyngbya infections that have been treated for either 3+ years, 2+ years, or 2 months, and were considered as separate populations for this trial. In addition to lyngbya collected from treatment sites, samples were also collected from an area outside of the current treatment program to serve as an additional treatment group to compare the response of lyngbya never receiving prior treatment to those samples collected from the previously described treatment sites. Lyngbya from this unexposed area was also used as a control site since it never received any algaecide treatment during the duration of this trial.

Treatment protocols followed the methods previously described and the treatment combination of Captain XTR® + AMP® was applied at maximum label rate to all treatment units, with an exposure time of 72 hours. For this trial, an evaluation of viability was performed 11-days post-treatment to assess lyngbya response to treatment and control. A series of two treatments were performed on each sample unit in consecutive order and occurred at 3-week intervals.

Lyngbya response to treatment was determined by evaluating the overall decrease in viability of lyngbya samples. To determine health of lyngbya prior to treatments, a pre-treatment viability level was obtained and compared to viably assessments conducted after the first and second treatments.

Results and Discussion

Algaecide Treatment Efficacy

All treatment combinations significantly decreased lyngbya viability 11-days post the third consecutive treatment exposure when compared to the untreated control ($p < .05$) (Table 2). Following the first treatment, only combinations utilizing Captain XTR® were significantly less viable than the untreated control ($p<.05$)(Table 2). Following the second consecutive treatment, lyngbya treated with combinations utilizing Captain XTR®, as well as, Cutrine Ultra® + AMP® were significantly reduced in viability when compared to the untreated control ($p<0.05$)(Table 2).

These results indicate that current algicide treatment protocols being utilized at Lake Gaston are effective at reducing lyngbya viability after consecutive treatments. It also indicates that Captain XTR® has the potential of producing a response in viability earlier in the treatment season and that efficacy does not appear to be directly impacted by the removal of AMP® from Captain XTR® treatment combinations (Table 2). However, it is important to note that these treatments are performed in a laboratory setting and do not incorporate all environmental conditions that could potentially impact efficacy if chelated copper algaecides are not applied in combination with AMP®.

Potential of Early Season Control

Significant reductions in lyngbya viability after exposure to all treatment combinations did not occur during either the February or March time periods ($p>0.05$)(Table 3). Response in viability levels to treatment combinations were found in April, but with high variability within each treatment combination (Table 3). The only chemical combination in April that produced significant reduction in viability when compared to the untreated control was Cutrine Ultra® + AMP® ($p<0.05$). Except for Captain XTR® alone, all treatment combinations produced significantly reduced viability levels in May when compared to the untreated control ($p<0.05$)(Table 3).

These results indicate that there is no potential of early season treatments for the Lake Gaston lyngbya treatment program, and April initiation of the annual season is the most appropriate in terms of efficacy (Table 3). Lyngbya showed no response to any treatment combination applied during the early season months (February and March) when water temperatures were below 65°F. Captain XTR®, Cutrine Ultra®, nor AMP® have a water temperature restriction contained in their label, therefore these results indicate that some unknown physiological response related to lyngbya could be the driver for efficacy in regards to these treatment combinations. It is interesting to note that during the months of February and March, a dark unknown organic compound was found in high densities and attached to the sheaths of lyngbya filaments (Figure 1). This organic material has not been documented in previous trials that occurred during the summer and early fall when water temperatures were above 65°F. In April and May, lyngbya began producing new growth which consisted of filaments that were not as robust in size or sheath thickness (Figure 1). This combination of decreased organic matter and increased new growth could help explain the timing of increased efficacy in algaecide treatments.

Impacts of Continual Annual Treatments

Overall, all treated sites displayed significantly reduced viability in lyngbya after the second consecutive treatment when compared to the untreated control ($p<0.001$) (Table 4). Sites that were actively being targeted by the lyngbya treatment program displayed significantly decreased viability following the initial treatment when compared to the untreated control (Table 4). Lyngbya from the site that was included into the program in 2024 and has only received 2 consecutive monthly treatments displayed the greatest variability in the level of viability reduction (Table 4). It should be noted that lyngbya viability at the initiation of treatments was significantly different among sites ($p<0.05$), with sites targeted during their first (2024) and third year (2021) showing significantly decreased viability than the sites that have not received previous treatments (control and never treated) (Table 4).

Temporal trends in lyngbya response to algaecide treatments could not be determined based on when a site was initiated into the lake-wide treatment program (Table 4). Although the response of lyngbya viability did not seem to correspond with the length of time a site was targeted by the program, all sites responded to treatments with no indications of decreased

efficacy. It is interesting to note that prior to treatment the overall viability of lyngbya from areas that were targeted by the Lake Gaston lyngbya treatment program were decreased from that of lyngbya collected outside of the program. These areas also displayed a greater response to treatments than lyngbya that had not received previous treatments. This could indicate that lyngbya from sites targeted by the program were in a state of decreased health from consecutive yearly treatments.

Conclusion and Management Implications

Overall, all three trials were able to provide further understanding of factors that influence lyngbya treatment programs. Results confirmed that treatment protocols, including treatment combinations and timing, were appropriate for the Lake Gaston lyngbya treatment program. Impacts from continual annual treatments were not detected, indicating that protocols do not need to be adjusted based on the length of time a site has been targeted by the program.

- For algicide treatment efficacy, results found that all treatment combinations provided significantly reduced viability in comparison to the control by the end of the third treatment cycle. Results also indicated that Captain XTR® has the potential of producing a response in viability earlier in the treatment season and that efficacy does not appear to be directly impacted by the removal of AMP® from Captain XTR® treatment combinations in the absence of external environmental factors.
- When looking at the potential for early season control, results found that lyngbya does not respond to any treatment combination in the months of February or March when water temperatures are below 65°F. These results indicate that there is no potential for providing lyngbya control during the colder, winter months.
- Responses in lyngbya viability did not seem to correspond with the length of time a site was targeted by the lyngbya treatment program and therefore efficacy was not impacted by continual annual treatments.

References

Bishop, W.M, B.E. Willis, and C.T. Horton. 2015. Affinity and efficacy of copper following an algicide exposure: application of the critical burden concept for *Lyngbya wollei* control in Lake Lay, AL. Environmental Management 55:983-990.

Calomeni, A.J., C.M.Kinley, T.D. Geer, M. Hendrikse, and J.H. Rodgers Jr. 2018. *Lyngbya wollei* responses to copper algaecide exposures predicted using a concentration-exposure time (CET) model: Influence of initial biomass. Journal of Aquatic Plant Management. 56: 73-83.

Lembi, C.A. 2000. Relative tolerance of mat-forming algae to Copper. Journal of Aquatic Plant Management 38: 68 - 70.

Peterson, M.A., A. Collavo, R. Ovejero, V. Shivrain, and M. Walsh. 2017. The Challenge of Herbicide Resistance Around the World: A Current Summary. Pest Management Science 74:2246-2259.

Richardson, R.J. 2008. Aquatic Plant Management and the Impacts of Emerging Herbicide Resistance Issues. Weed Technology 22: 8-15.

Rodgers, J.H. and M.L. Pietruszewski. 2021. Ecology and management of algae and harmful algal blooms. Pages 101 – 112 *in* L.A. Gettys, W.T. Haller, and D.G. Petty, editors. Biology and Control of Aquatic Plants. Aquatic Ecosystem Restoration Foundation, Marietta, Georgia.

Tables

Table 1. Overview of experimental set-ups for each of the three trials used to assess aspects of lyngbya treatment programs.

Overview of Trials Used to Assess Lyngbya Treatment Program							
	Exposure Time (hrs)	Evaluation Period (days)	Number of Consecutive Treatments	Captain XTR®	Captain XTR® + AMP®	Cutrine Ultra®	Cutrine Ultra® + AMP
Algaecide Treatment Efficacy	72	11	3	X	X	X	X
Potential for Early Season Control	48	10	0	X	X	X	X
Impacts of Continual Annual Treatments	72	11	2		X		

Table 2. Changes in percent viability of lyngbya in response to a series of three sequential algaecide treatments using a combination of Captain XTR® + AMP®, Cutrine Ultra® + AMP®, as well as, Captain XTR® and Cutrine Ultra® independently. DAT # indicates if results are following the first, second, or third consecutive treatment exposure. All analysis shown compares viability responses following each consecutive exposure for treatment combinations individually. Means within the same row followed by the same letter do no significantly according to Tukey's HSD ($p<0.05$).

Lyngbya Viability – Algaecide Treatment Efficacy					
	Control	Captain XTR®	Captain XTR® + AMP®	Cutrine Ultra®	Cutrine Ultra® + AMP®
DAT #1	91 *a*	75 *b*	75 *b*	82 *ab*	85 *ab*
DAT #2	91 *a*	69 *b*	62 *b*	75 *ab*	65 *b*
DAT #3	95 *a*	59 *b*	56 *b*	65 *b*	66 *b*

Table 3. Changes in percent viability of lyngbya in response to algaecide treatments using a combination of Captain XTR® + AMP®, Cutrine Ultra® + AMP®, as well as, Captain XTR® and Cutrine Ultra® independently during treatments occurring in February, March, April, and May. Monthly water temperatures were 55°F in February, 60°F in March, 70°F in April, and 75°F in May. All analysis shown compares viability responses for treatment combinations individually within a single month. Means within the same row followed by the same letter do no significantly according to Student's t-test ($p<0.05$).

Lyngbya Viability – Potential for Early Season Control					
	Control	**Captain XTR®**	**Captain XTR® + AMP®**	**Cutrine Ultra®**	**Cutrine Ultra® + AMP®**
February	**94 *a***	**90 *a***	**95 *a***	**94 *a***	**89 *a***
March	**93 *a***	**92 *a***	**94 *a***	**90 *a***	**91 *a***
April	**94 *a***	**85 *a***	**78 *a***	**63 *ab***	**62 *b***
May	**92 *a***	**81 *ab***	**74 *b***	**77 *b***	**74 *b***

Table 4. Changes in percent viability of lyngbya in response to a series of two sequential algaecide treatments using a combination of Captain XTR® + AMP®. Samples were collected from areas that have been receiving continual treatments by the Lake Gaston lyngbya treatment program for either 1, 3, or 4 years and compared to samples collected from an area outside of the treatment program. All analysis shown compares viability responses following each consecutive exposure for the level of continual treatment individually. Means within the same row followed by the same letter do no significantly according to Student's t-test ($p<0.05$).

Lyngbya Viability – Impacts of Continual Annual Treatments					
	Control	**Never Treated**	**Year 1**	**Year 3**	**Year 4**
Pre-Treatment	**95 *a***	**93 *a***	**60 *b***	**65 *b***	**73 *ab***
DAT #1	**91 *a***	**79 *a***	**44 *b***	**33 *b***	**53 *b***
DAT #2	**93 *a***	**59 *b***	**20 *c***	**6 *c***	**19 *c***

Figures

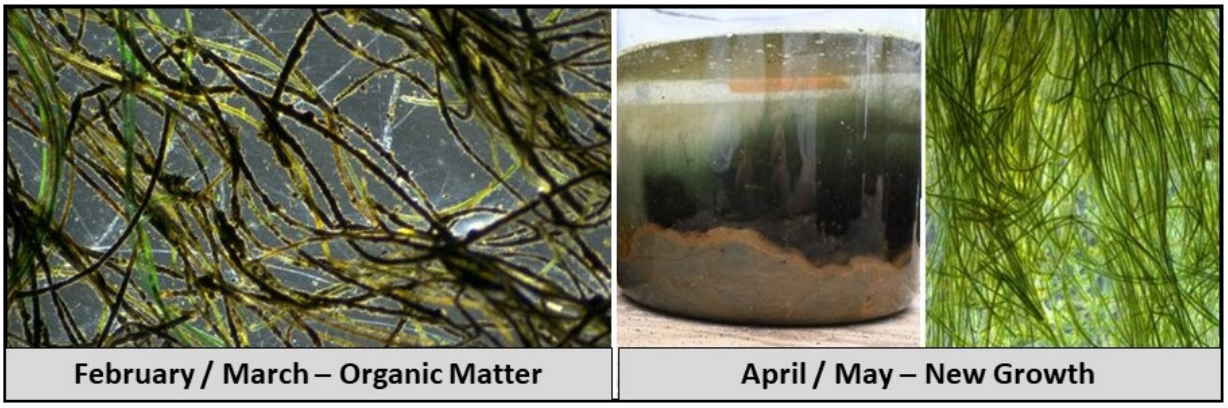


Figure 1. Images of organic material observed on lyngbya during early season treatments and new growth that began forming in April and May.