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Aquatic Nuisance Species Research Program

Pilot-Scale Optimization

Research on Algae Flotation Techniques (RAFT)

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Abstract

The impacts of harmful algal blooms (HABs) on US national waterways continue to cause significant economic and environmental damage. Researchers at the US Army Engineer Research and Development Center successfully demonstrated the *Research on Algae Flotation Techniques* (RAFT) project at pilot scale. This study was designed to show that the surface concentrations of algal biomass can be effectively increased with near linear scalability utilizing the natural methods by which some algae entrap air within excreted mucilage for flotation. The surface concentration of cyanobacteria measured as phycocyanin pigment increased by six-fold after RAFT flocculation treatment. Further optimization of chemical delivery systems, mixing, and dissolved air exposure will be required before full scale readiness.

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Preface

This study was conducted for US Army Engineer Research and Development Center (ERDC) Aquatic Nuisance Species Research Program (ANSRP) under AMSCO code 008284, "Pilot Scale Optimization—Research on Algae Flotation Techniques (RAFT)."

The work was performed by the Emergency and Operational Support Branch of the Research and Engineering Division, US Army ERDC–Construction and Engineering Research Laboratory (CERL). At the time of publication, Ms. Ellen Hartman was branch chief; and Dr. George Calfas was division chief. The deputy director of ERDC-CERL was Ms. Michelle Hanson, and the director was Dr. Andrew Nelson. The work was also performed by the Aquatic Ecology and Invasive Species Branch of the Ecosystem Evaluation and Engineering Division, US Army ERDC–Environmental Laboratory (EL). At the time of publication, Mr. Alan Katzenmeyer was branch chief; and Mr. Mark Farr was division chief. The deputy director of ERDC-EL was Dr. Brandon Lafferty, and the director was Dr. Edmund Russo. The work was also performed by the USACE St. Louis and Jacksonville districts.

COL Christian Patterson was commander of ERDC, and Dr. David W. Pittman was the director.

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1 Introduction

1.1 Background

Algae harvesting has gained considerable attention in recent years because of its potential as a renewable source for biofuel. In many cases, advanced growth systems are used to culture algae under controlled conditions to achieve optimized algal growth (Milledge and Heaven 2013; Slade and Bauen 2013). However, recent studies have investigated the ability to harvest algae from natural waters to obtain a resultant biomass suitable as feedstock for fuel and plastics (Chia et al. 2020; Page et al. 2020). To date, optimization of open water algae harvesting is warranted, as the most critical steps in this process are polymer selection and algae dewatering method. Flocculant selection may present challenges when the agents are introduced into the environment as many high performing synthetic polymers are typically applied in closed systems and may potentially subject aquatic organisms to toxicity. Thus, the use of biopolymers as flocculants are preferred. Not only are biopolymers more environmentally friendly but are also a more attractive option to obtain a cleaner algal feedstock.

1.2 Objectives

To obtain maximum algal flocculation, factors such as algae species, polymer type, and charge are to be considered (Pugazhendhi et al. 2019). Microalgae can exist in single cell, filamentous, or colony form with the ability to aggregate and produce extracellular polymeric substances (EPS) or mucilage. Because cell wall composition of microalgae (polysaccharides, proteins, and lipids) contributes to its negative surface charge, charge neutralization and electrostatic bridging are important mechanisms for algae destabilization and flocculation (Speranza et al. 2022). Natural polysaccharides, such as xanthan gum, carries a net negative charge and may moderately bind to the gelatinous sheet of cyanobacteria if present in abundance. At present, published data reporting the effect of cationic and anionic polymers added either sequentially or simultaneously is limited. Findings from preliminary jar tests show that xanthan gum (XG) can be a secondary or tertiary additive in the chemical crosslinking of cationic flocculants to further aid in the aggregation of algal colonies. Cender et al. (2023) investigated the anionic polysaccharide as a means to exploit the natural flocculation mechanisms of algae. Findings from this study suggest that XG can significantly increase the efficacy of algae flotation upon interaction with flocs formed by cationic polymers. In this manner, lower concentrations of synthetic polymers may be incorporated with biopolymer applications to enhance flocculation efficiency.

In addition to polymer selection, another consideration is the modification of dissolved air flotation (DAF) treatment methods to yield varying outcomes in algal flotation. Nano- and microbubbles interact differently with suspended particles based on physical and surface chemistry properties (Rahmana et al. 2014). Findings from Ushikubo et al. (2010) suggest that nanobubbles are capable of remaining stable in water for long periods of time (hours to days). Nanobubbles are also noted for their efficiency in water treatment to carry suspended solids to the water surface (Movahed et al. 2021).

1.3 Approach

This study aimed to flocculate and float algae to the water surface at pilot scale evaluating DAF treatment by nano- and microbubbles. Using a biopolymer and a cationic coagulant as the primary and secondary agents, respectively, this study was conducted for the purpose of (1) evaluating differences in DAF by nano- and microbubbles, (2) obtaining reproducibility of results observed from jar tests under laboratory conditions, and (3) determining scalability of the treatment process.

2 Materials and Methods

2.1 Materials

A mixed species algae culture containing cyanobacteria and Chlorophyta was collected from an environmental test pond in Champaign, Illinois. XG, acting as a simulated EPS, and cationic Polyacrylamide (cPAM), the electrostatic coagulant and supplementary EPS, were purchased from Chemical Systems of Orlando, Inc. The dissolved air flotation unit system was purchased from Moleaer, Inc. Concrete outdoor test ponds utilized for algal treatments were 5.56 m × 5.56 m × 1.7 m (14,000 US gal.) each.* A Van Dorn water sampler, purchased from Grainger, was used to collect water samples below the water surface. An In-Situ Aqua Troll 600 multiparameter data sonde was used to measure water quality parameters including algal pigment fluorescence. The Appendix contains details regarding the experimental setup and equipment.

2.2 Preparation of outdoor test ponds

Algae samples were cultured in BG11 media under laboratory conditions at approximately 23°C. Outdoor test ponds, located at a test facility in Vicksburg, Mississippi, were stocked with subcultures of algae during early summer. The algae cultures were allowed an acclimation period of 14 days in the outdoor ponds before testing. After algae reached the stationary growth phase, water quality parameters were recorded before treatment to obtain baseline measurements in the test ponds. Background samples were also collected before treatment. Four test ponds were prepared for algae treatments. Two of the four test pond cultures failed leaving Ponds 1 and 2 for treatment with nano- and microbubbles, respectively.

2.3 Algae treatment

Upon obtaining desired algal concentrations in the test ponds, XG and cPAM were dosed into the ponds, both at concentrations of 5 mg/L, respectively. The chemicals were incorporated into the ponds upstream of the DAF unit which also functioned as a mixing mechanism for the polymers. Flocculant ratios and concentrations were previously determined

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* For a full list of the spelled-out forms of the units of measure used in this document, please refer to US Government Publishing Office Style Manual, 31st ed. (Washington, DC: US Government Publishing Office, 2016), 248–52, https://www.govinfo.gov/content/pkg/GPO-STYLE-MANUAL-2016/pdf.

from iterative preliminary experiments reported in Cender et al. (2023). The pilot scale DAF unit was used for the purpose of subjecting algae flocs to micro- and nanobubbles in the test ponds. The DAF unit consisted of a 2 in. diameter inlet and outlet, a proprietary nanobubble generation cell, a 3/4 horsepower flow pump, and an air compressor. The test ponds were treated with the XG via the DAF mechanism for mixing and delivery for 20 min before cPAM was introduced into the system. The nano- and microbubbles were generated using test pond source water with the treatment mixture present. Flotation was recorded as relative fluorescence units (RFU) starting after the cPAM was added (40 min–60 min). Pressurized, algae-laden water was circulated into the ponds at a depth of approximately 4.5 ft to 5 ft. Reaction rates were monitored with data sondes during the treatment process to measure real time changes in water phycocyanin levels. Van Dorn water samplers were used to collect samples from both test ponds at 5 ft to determine biovolume and cell density measurements before and after treatment.

2.4 Sample analysis

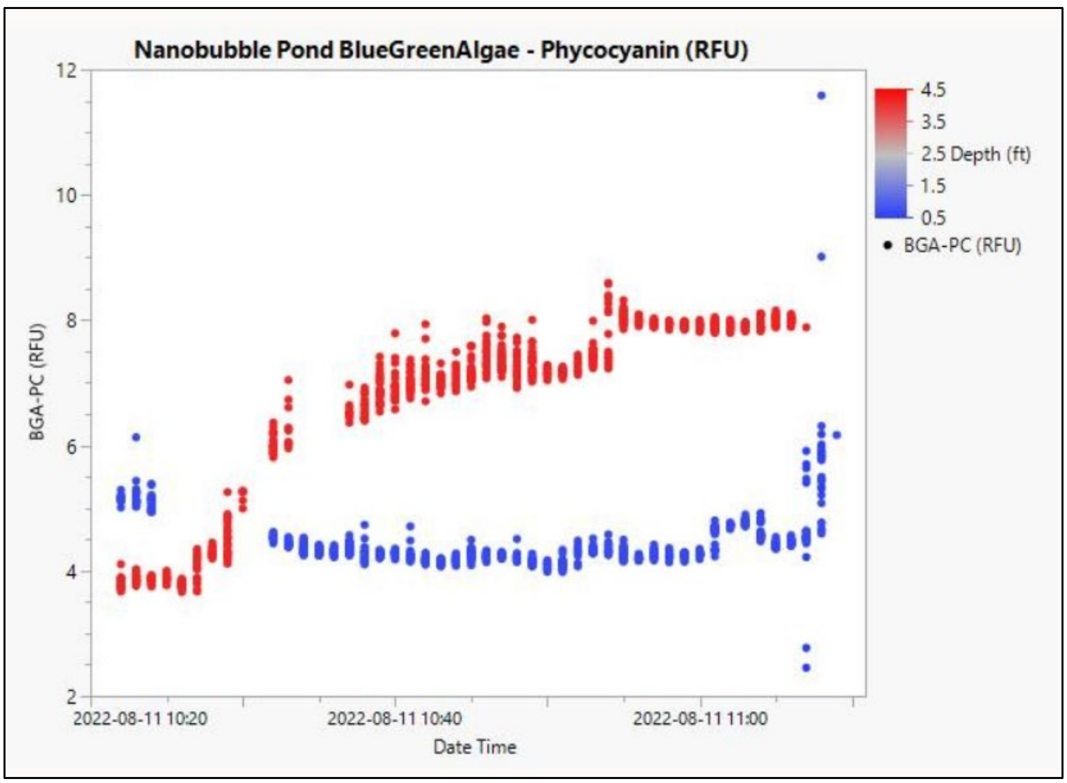
Water quality parameters were measured on-site using multiparameter data sondes and a Hach™ DR1900 spectrophotometer with Test 'N Tube test reagents. The parameters measured were phycocyanin (to indicate cyanobacteria), temperature, total nitrogen, total phosphorus, and chemical oxygen demand. Samples collected before and after treatment were analyzed by a third-party test lab for microbial identification, cell count, and total biovolume.

3 Results and Discussion

3.1 DAF (dissolved air flotation) by nanobubbles

Phycocyanin levels after DAF nanobubble infusion increased from 4.1 RFU to 8.0 RFU at 4.5 ft while surface concentrations decreased from 5.3 RFU to 5.0 RFU after 50 min (Figure 1). Surface phycocyanin levels were overall considerably lower than the values observed at 4.5 ft. The observed shift is indicative of preferential sedimentation of cyanobacteria flocs upon interaction with nanobubbles, although the effect of sedimentation was relatively minimal. Nanobubbles are known to be far more stable and prone to Brownian motion than microbubbles. The nanobubbles did not appear to have the buoyant force necessary to carry the algae flocs to the surface and the flocs that gained enough new mass drifted downward. No significant flotation of algal flocs was observed on the water surface after 60 min post treatment.

Figure 1. Phycocyanin concentrations near the water surface (0.1 ft–0.5 ft) and at 4.5 ft after dissolved air flotation (DAF) nanobubble infusion. After treatment, phycocyanin levels indicate that cyanobacteria exhibited higher rates of sedimentation rather than flotation.



The most abundant organisms present in the test pond source were Chlorophyta (green algae) in terms of cell density or count in cells per liter (Figure 2). The concentrations of these low biovolume cells dramatically increased post-nanobubble treatment likely because of the effects of the treatment and concentrated biomass formation. The total biovolume of cells is indicative of cell volume rather than cell abundance as with the density metric. The Chlorophyta, despite being much smaller in individual cell volume, dominate the total biovolume after treatment (Figure 3). The data suggest that cyanobacteria flocs may have undergone a minimal degree of flotation, though it is uncertain if this is because of nanobubbles, noise, or error. The biomass did not accumulate significantly near the water surface during the length of the experiment. Chlorophyta exhibited the highest magnitude of sedimentation as indicated by density and biovolume (Figure 2 and Figure 3).

Figure 2. Cell density of Bacillariophyta (diatoms), Chlorophyta (green algae), and cyanobacteria (blue-green algae) after DAF nanobubble infusion at 4.5 ft below water surface.

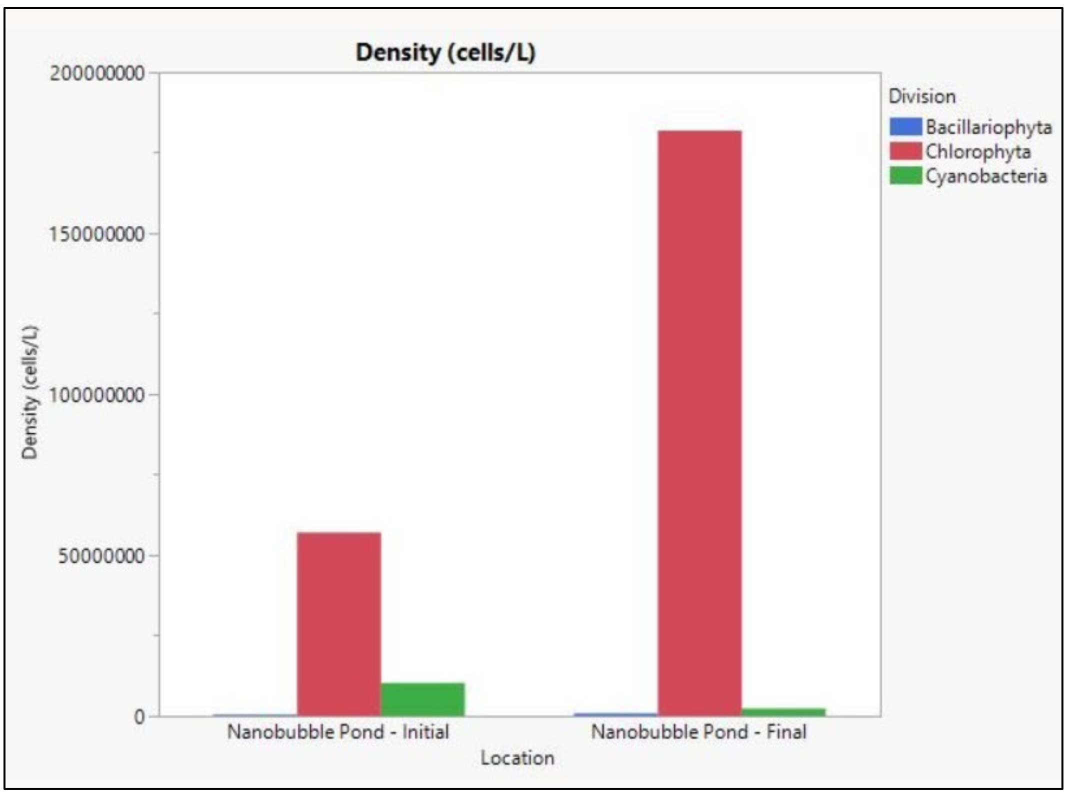
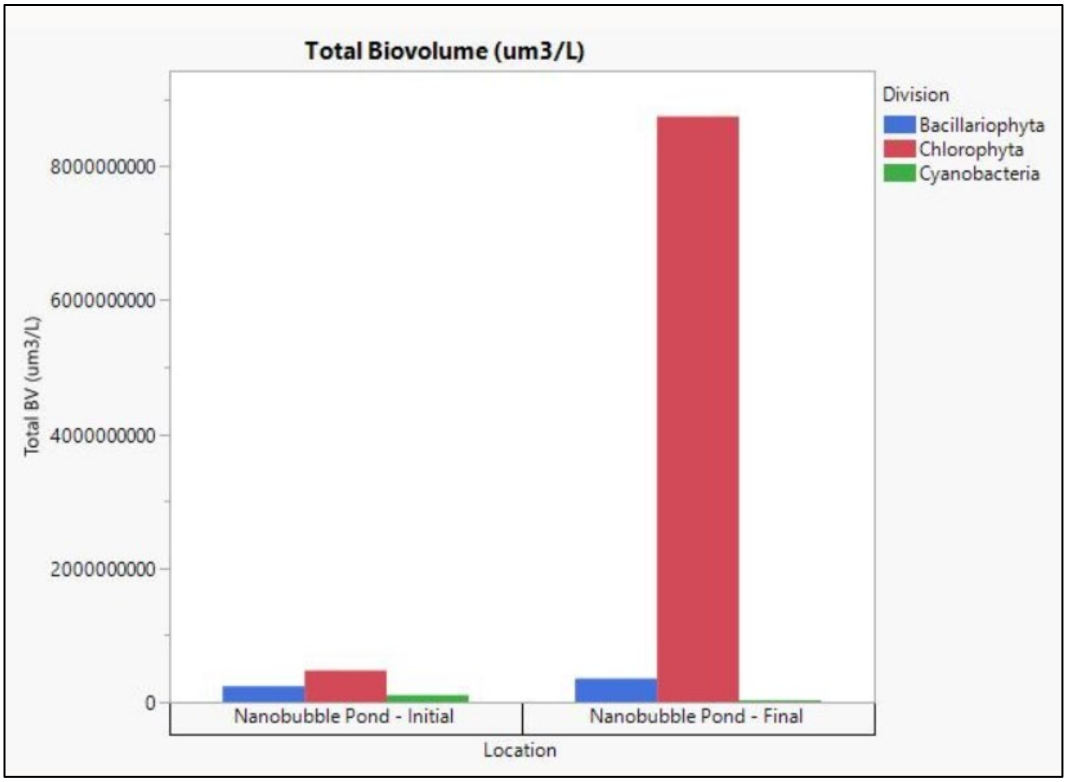


Figure 3. Biovolume of Bacillariophyta (diatoms), Chlorophyta (green algae), and Cyanobacteria (blue-green algae) after DAF nanobubble infusion at 4.5 ft below water surface.



3.2 DAF by microbubbles

Following DAF microbubble infusion, phycocyanin levels at 4.5 ft remained relatively constant throughout the 40 min evaluation period in which the concentration modestly decreased from 4 RFU to 3 RFU (Figure 4). Near the surface, phycocyanin levels increased from 5 RFU to 30 RFU with an abbreviated spike observed after 25 min at 36 RFU (Figure 4). Contrary to results observed after nanobubble exposure, algal flocs of a foam consistency became visible on the water surface following microbubble treatment (Figure 5).

The cell density of Chlorophyta present before and after the DAF microbubble treatment demonstrated change likely indistinguishable from measurement noise. The cell density of measured cyanobacteria was more abundant after treatment with uncertainty about whether this was caused by noise or a lack of adequate exposure to microbubbles after flocculation. The cell density of cyanobacteria increased from 7.6×10^6 cells/L to 2.6×10^7 cells/L (Figure 6).

Figure 4. Near surface (0.1 ft–0.5 ft) and below surface (4.5 ft) phycocyanin concentrations after DAF microbubble infusion. Increased phycocyanin concentrations near water surface indicates flotation of algae flocs 40 min after treatment.

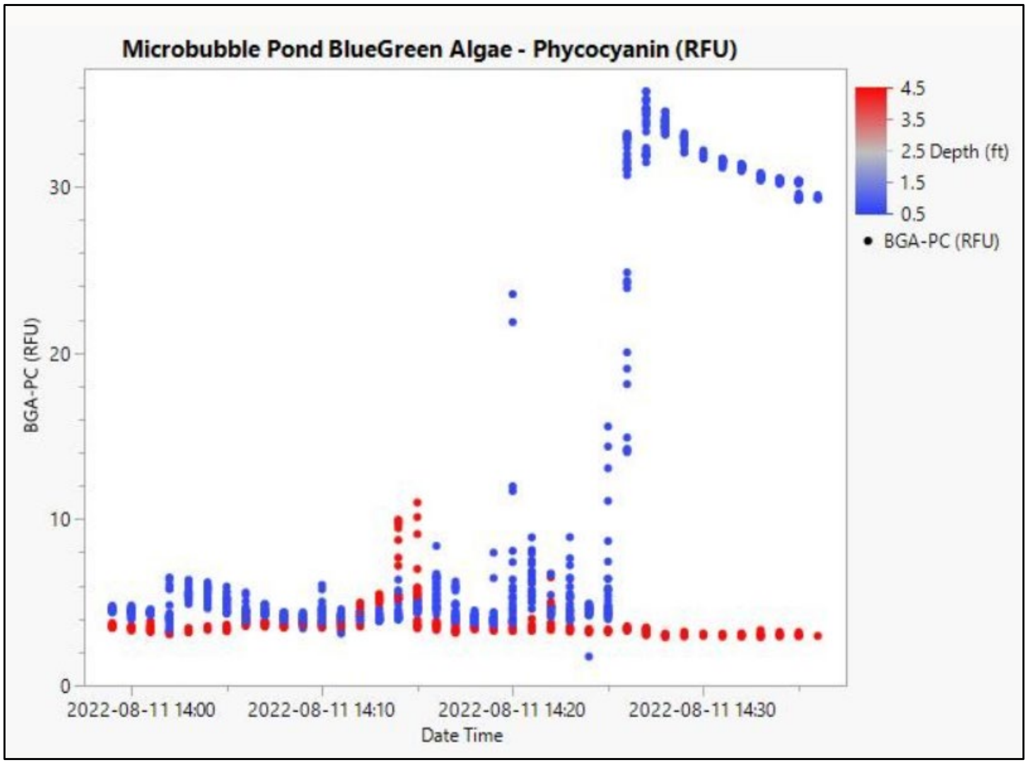
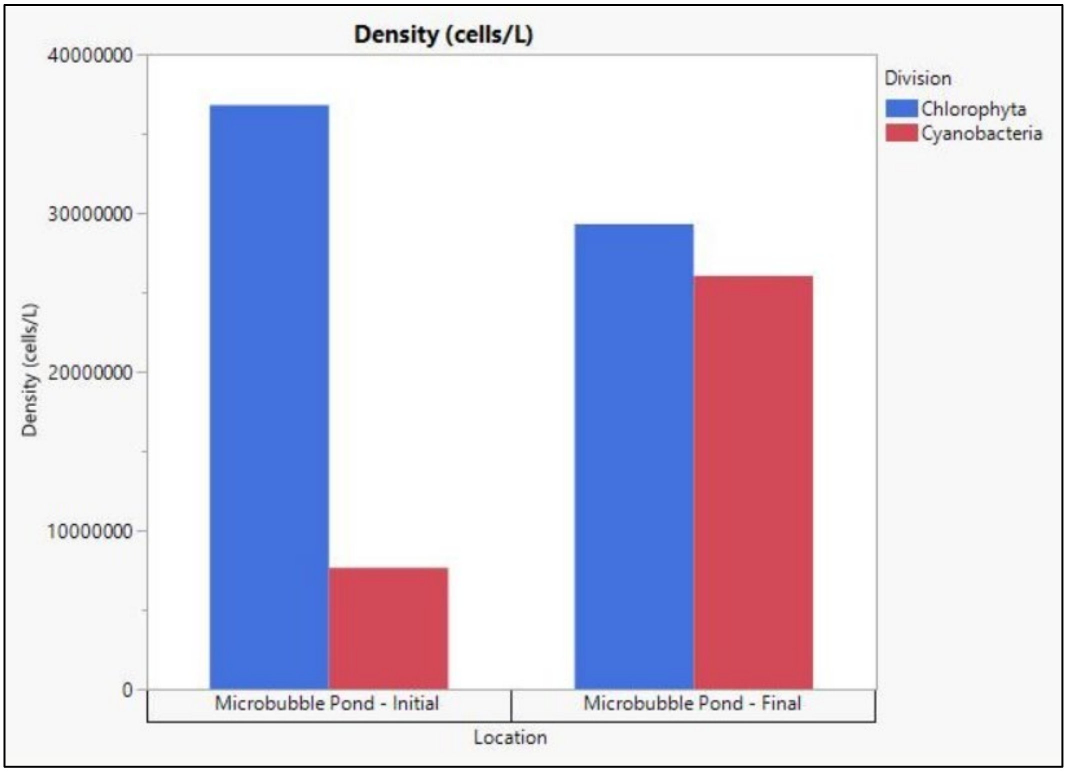


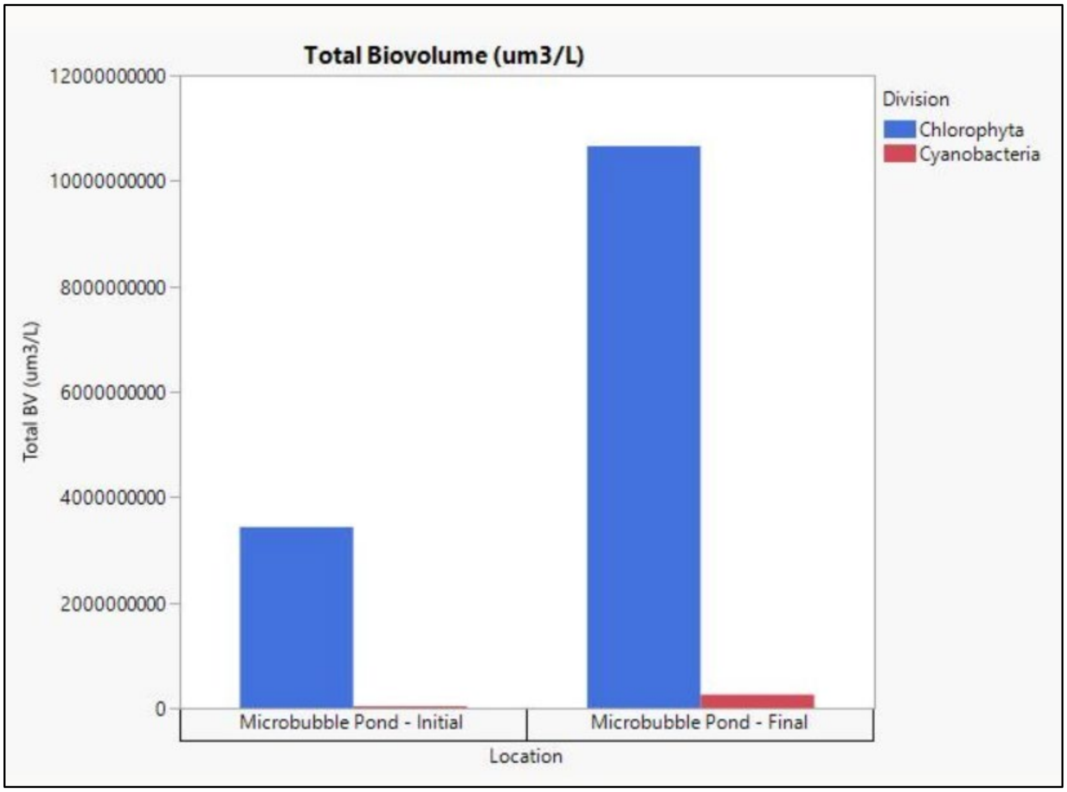
Figure 5. Surface accumulated biomass after chemical and DAF microbubble treatment.

Figure 6. Cell densities for Chlorophyta and Cyanobacteria at 4.5 ft after microbubble treatment. Increase in cyanobacteria posttreatment is consistent with biovolume, though the cell density of Chlorophyta at 4.5 ft declined posttreatment.



The total biovolume of Chlorophyta at 4.5 ft increased after the 30 min evaluation period from 3.5×10^9 µm^3/L to 10.5×10^9 µm^3/L. A minimal increase in cyanobacteria biovolume was observed after 30 min following DAF microbubble infusion at 4.5 ft (Figure 7). These data suggest that the chemical treatment and microbubbles will not offer complete removal of microalgae in the system as both predominant organism classifications remained after treatment with minimal significant change (Figure 6 and Figure 7). The dramatic increase in surface phycocyanin and the observed accumulation of surface biomass is indicative of significant biomass capture, however (Figure 4 and Figure 5).

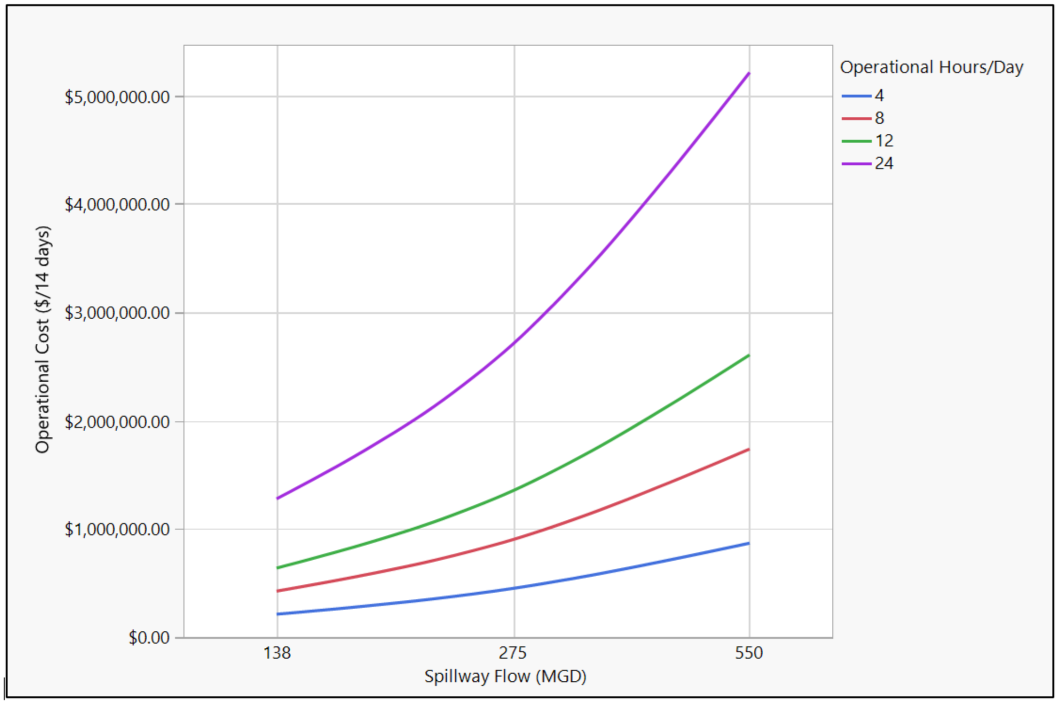
Figure 7. Total biovolume of Chlorophyta and Cyanobacteria at 4.5 ft after DAF microbubble infusion. Concentration increases by both divisions is indicative of floc precipitation.



3.3 Scalability

The research team used quantitative metrics to determine scalability of the economic scalability of operating a rapid algae flotation technique (RAFT) treatment system. The estimated capital cost of the system is in the range of $600 k according to informal market research and 2021 cost estimates for similar DAF water treatment systems, chemical storage and delivery, and peripherals (Page et al. 2021). The most significant recurring cost of the system is projected to be consumable chemical and operational costs that will be scaled based on system uptime. Using a spillway as a use case example, a 14 day system operated at 8 hr per day could require just under $1–$1.5 M in operational costs during average spillway flow (Figure 8).

Figure 8. Estimated operational cost of a system deployed for 14 days on a spillway. The estimate does not include labor, fuel, or capital investment.



4 Conclusions

The key outcomes of this RAFT pilot demonstration are,

- the chemical treatment and algae flocculation was consistent with laboratory scale experiments suggesting linear scalability and met performance objectives,
- nanobubble exposure to flocculated algae had no observable significant short-term effects and long-term effects were not included in this study,
- microbubble exposure to flocculated algae created significant surface biomass flotation, and
- the treatment may be best suited for partially enclosed waterways such as canals upstream of a spillway rather than in open water.

Challenges to be addressed are,

- chemical delivery, mixing, and DAF exposure to the flocculated algae becomes increasingly complex with scale and requires optimization,
- microbial identification data suggest some biomass sedimentation, which will require optimization, and
- no definitive conclusion has been made on taxonomic selectivity and will need to be investigated.

The 2022 RAFT pilot demonstration was a critical step in understanding the knowledge gaps and optimizations necessary to bring the system to the next technology readiness level. The data collected from the study indicate that the RAFT process performs as expected at larger scales. The demand on raw materials is linear and therefore predictable and scalable. However, challenges with adequate chemical delivery, mixing, and dissolved air exposure to the flocs will require additional investigation and optimization before a system could operate to full effect. In the short term, RAFT techniques can be utilized to supplement existing microbubble DAF systems to improve biomass flocculation and surface concentrations. Further understanding and optimization of the mixing and fluid dynamics of the algal suspension may lead to a sixfold increase in surface concentration of phycocyanin responsive algae.

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Appendix: Experimental Components

Table A-1. Equipment and test facilities.

Concrete Test Pond Dimensions; L × W × D (ft)	5.56 m × 5.56 m × 1.7 m (14,000 US gal.) Each
Nanobubble/microbubble generator	Moleaer Clear 50 50 GPM Gas feed range 60–100 (psig) ¾ HP pump Compressor draw 0.875 HP Inlet/outlet 2” in.

Table A-2. Algae growth media.

Chemical Name	CAS#
Potassium nitrate	7757-79-1
Ammonium nitrate	6484-52-2
Urea phosphate	4861-19-2
Boric acid	10043-35-3
Copper sulfate	7758-98-7
Iron Ethylenediaminetetraacetic acid (EDTA)	15708-41-5
Manganese sulfate	7785-87-7
Sodium molybdate	7631-95-0
Zinc sulfate	7733-02-0

Table A-3. Algae composition.

Chlorophyta	Cyanobacteria
cf. *Cyclotella* sp.	cf. *Chroococcus* sp.
cf. *Chlorella* sp.	cf. *Phormidesmis* sp.
Chlorella vulgaris	*Planktolyngbya limnetica*
Chloroidium ellipsoideum	cf. *Planktolyngbya* sp.
cf. *Raphidocelis* sp.	*Pseudanabaena* sp.
Desmodesmus spp.	cf. *Pseudanabaena* sp.
Monoraphidium contortum	—
Monoraphidum pusillum	—
Monoraphidium tortile	—
Pandorina sp.	—
Tetradesmus sp.	—

Table A-4. Weather data for 2022—Vicksburg, Mississippi.

Date	Temp High/Low (°F)	Rain Yes/No (Y/N)
25 July	86/73	Y
26 July	89/75	Y
27 July	88/75	N
28 July	89/75	N
29 July	89/75	N
30 July	76/73	N
31 July	87/73	N
01 Aug	85/73	Y
02 Aug	85/72	N
03 Aug	82/72	Y
04 Aug	84/73	N
05 Aug	84/75	Y
06 Aug	87/72	N
07 Aug	89/73	N
08 Aug	90/75	Y
09 Aug	83/72	Y
10 Aug	78/72	N
11 Aug	82/72	N

Table A-5. Test pond parameters pretreatment.

Pond	Replicate	Chemical Oxygen Demand (mg/L)	Total Phosphorus (mg/L)	Total Nitrogen (mg/L)
1	1	187	5.02	17.1
1	2	236	5.3	26.9
1	3	196	5.33	17
2	1	493	8.9	25.6
2	2	420	4.71	11.1
2	3	418	7.74	23.2
3	1	291	4.45	10
3	2	256	4.33	17.5
3	3	264	4.47	11.1
4	1	451	8.86	32.4
4	2	428	10.3	10
4	3	402	9.01	15.7

Abbreviations

cPAM	Cationic Polyacrylamide
DAF	Dissolved air flotation
EDTA	Ethylenediaminetetraacetic acid
EPS	Extracellular polymeric substances
ERDC	Engineer Research and Development Center
HAB	Harmful algal bloom
RAFT	Research on Algae Flotation Techniques
RFU	Relative fluorescence units
XG	Xanthan gum

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14. ABSTRACT
The impacts of harmful algal blooms (HABs) on US national waterways continue to cause significant economic and environmental damage. Researchers at the US Army Engineer Research and Development Center (ERDC) successfully demonstrated the *Research on Algae Flotation Techniques* (RAFT) project at pilot scale. This study was designed to show that the surface concentrations of algal biomass can be effectively increased with near linear scalability utilizing the natural methods by which some algae entrap air within excreted mucilage for flotation. The surface concentration of cyanobacteria measured as phycocyanin pigment increased by six-fold after RAFT flocculation treatment. Further optimization of chemical delivery systems, mixing, and dissolved air exposure will be required before full scale readiness.

15. SUBJECT TERMS
Algae; Dissolved air flotation; Flocculation; Microbubble; Nanobubble; Polysaccharide; Xanthan gum

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