

**Project Title: Nitrogen and Oxygen Management Using Innovative Technologies to Improve Water Quality by BlueGreen Water Technologies****EXECUTIVE SUMMARY**

BlueGreen U.S. Water Technologies, Inc. (BlueGreen) is a leading global WaterTech company with an innovative suite of products designed for scalable treatment to improve water quality conditions and remediate harmful algal blooms in natural and man-made waterbodies, irrespective of size or shape. BlueGreen's proven technology successfully remediated waterbodies through various commercial applications across the United States of America, Israel, South Africa, Russia, and China, with activities coming soon in Ecuador and Colombia. BlueGreen presents this proposal to provide the City of Marco Island, Florida our approach to reducing nitrogen concentrations and increasing dissolved oxygen concentrations using our product, Lake Guard® Oxy, in nutrient-impaired surface waters around the city. The Lake Guard® Oxy is approved by the U.S. EPA, Organic Materials Review Institute (OMRI) listed, certified by the NSF/ANSI/CAN-60 for treatment in drinking water, and registered with the Florida Department of Agriculture and Consumer Services. The ready-to-use Lake Guard® products can be broadcasted manually from the shore, a boat, or areal dispersal by a licensed applicator.

**COMPANY BACKGROUND**

BlueGreen was founded in 2014 and began globally commercializing its Lake Guard® technology in 2017. BlueGreen is a privately-owned company based in Israel, with subsidiaries in the United States and in China. The science team comprises accomplished personnel with years of interdisciplinary expertise in business and science, including but not limited to water chemistry, physics, numerical modelling, phycology, microbiology, hydrology, aquatic and wildlife ecology, geospatial, and artificial intelligence.

**PROPOSED TECHNOLOGY****Lake Guard® Oxy Product**

The product, Lake Guard® Oxy, is a proprietary formulation that is comprised of 98% (w/w) sodium percarbonate that, when mixed with water, releases hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and 2% (w/w) of a food-grade biodegradable encapsulating agent. The dissolution of H<sub>2</sub>O<sub>2</sub> produces water and oxygen molecules and does not contain heavy metals or pollutants that could significantly harm the natural environment. H<sub>2</sub>O<sub>2</sub> has advantages as a supplemental oxygen source. This eco-friendly product was specially formulated to float and time-release and leave no trace or bioaccumulate in the environment.

**RATIONALE****Lake Guard® Oxy Technology to Produce Oxygen**

As Lake Guard® Oxy mixes with water, the resulting chemical reaction produces hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) is a strong oxidant that occurs naturally in aquatic systems, and quickly decomposes into water and gaseous oxygen (Figure 1.; Drábková et al., 2007). Additionally, and opportunistically, as H<sub>2</sub>O<sub>2</sub> decomposes, it releases hydroxyl radicals which are strong reactive oxygen species (ROS) that can be detrimental to the growth and function of Cyanobacteria and are capable of significantly reducing Microcystis and Planktothrix biomasses (Barrington et al., 2011;

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Matthijs et al., 2012). ROS act as a secondary messenger in biological processes such as response to environmental stresses. At high concentrations, ROS is very harmful to certain algae cells leading to oxidative damage to various cellular components comprising cell membranes, proteins and lipids (Figure 2; Rezayian et al. 2019). This oxidative stress then creates a chain reaction of cell death among colonial populations of harmful algae. Microcystis, for example, is an order of magnitude more sensitive than multiple species of green algae and diatoms (Drábková et al., 2007).

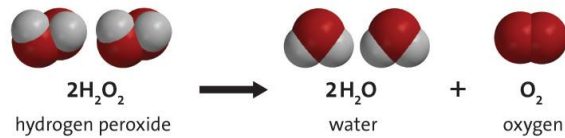


Figure 1. Simplified decomposition of hydrogen peroxide (Credit: Socratic.com).

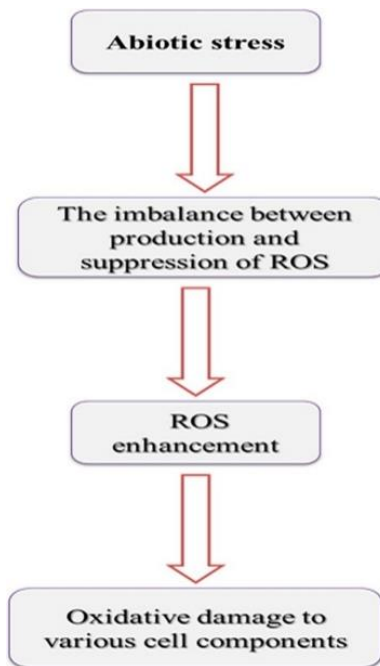


Figure 2. Oxidative damage and antioxidative system in algae graphical abstract (Credit: Rezayian et al. 2019).

### Lake Guard® Oxy Technology to Reduce Nitrogen

According to the ERD Nutrient Source Evaluation Report (2021), reuse irrigation contains elevated concentrations of both nitrogen (N) and phosphorus (P) and current N loadings to waterways in and around the City of Marco Island are excessive and enhancing an existing water quality impairment (Figure 3). In support of our proposed use of BlueGreen’s Lake Guard® Oxy, studies have shown that as hydrogen peroxide dosing concentration increases, concentrations of nitrate in the effluent decreases substantially due to stimulated plant growth and plant uptake of nitrate instead of ammonia. For instance, dosing hydrogen peroxide to constructed wetlands extended ammonia

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removal via nitrification to total inorganic nitrogen removal by nitrification and stimulated plant uptake of nitrate (Dinakar et al. 2020). Similar results could be beneficial to existing submerged aquatic vegetation as documented by Humiston & Moore Engineers' 2023 Tigertail Lagoon/Sand Dollar Island Ecosystem Restoration Project Proposal.

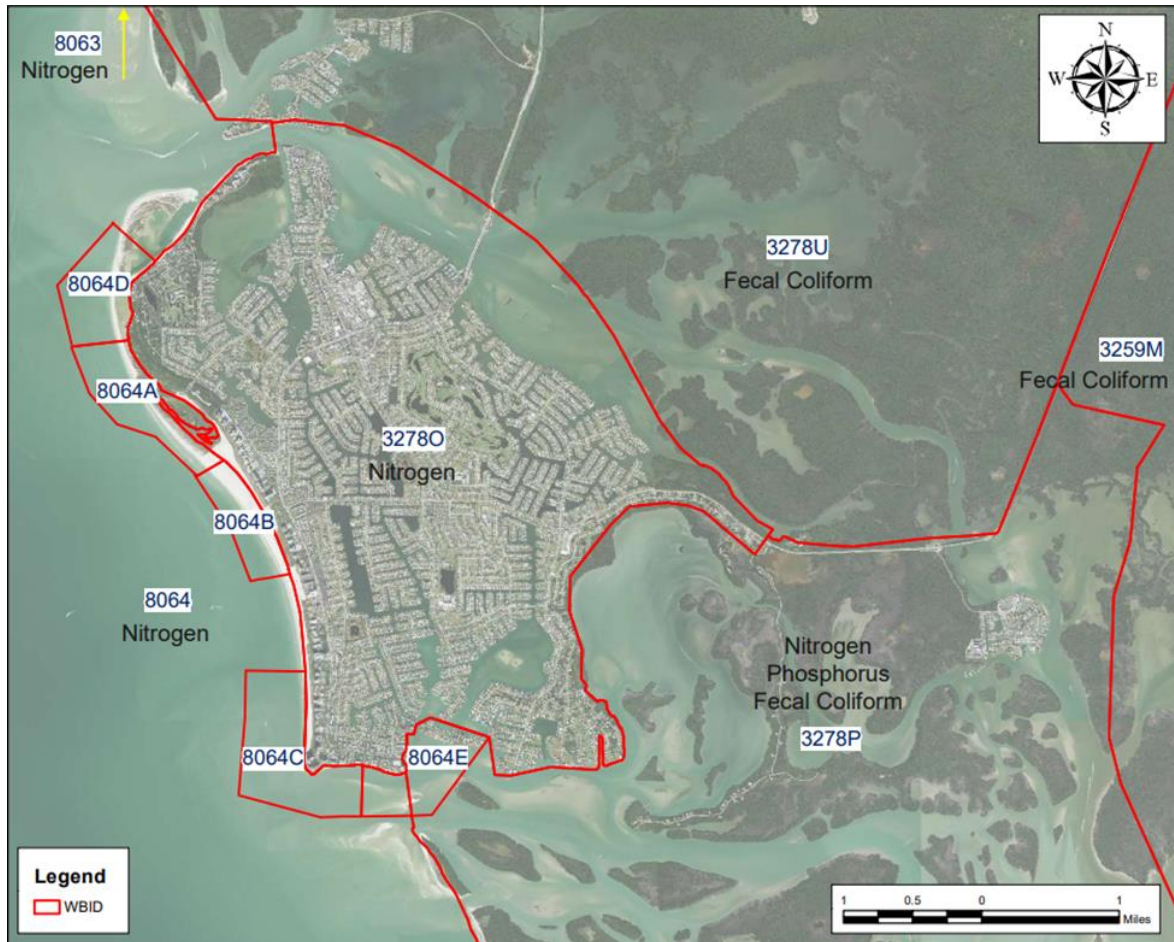


Figure 3. Unique Waterbody Identification Number (WBID) for the City of Marco Island, Florida waterways (Credit: ERD Nutrient Source Evaluation Report 2021).

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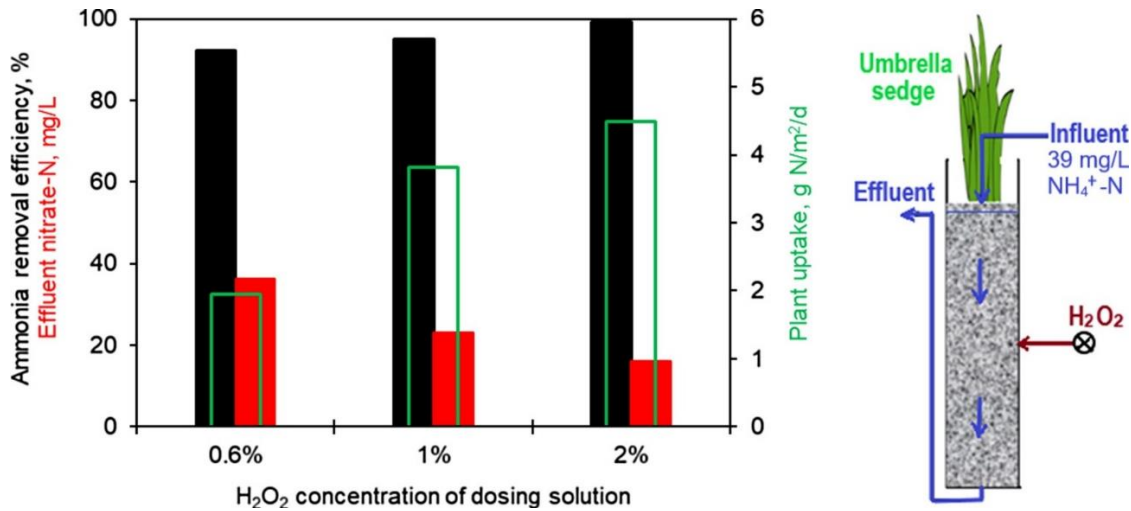


Figure 4. *Stimulating Nitrogen Biokinetics with the Addition of Hydrogen Peroxide to Secondary Effluent Biofiltration* (Credit: Dinakar et al. 2020).

Although tertiary wastewater treatment could provide a reliable source of water for reuse, ammonium ( $\text{NH}_4^+$ ) and nitrite ( $\text{NO}_2^-$ ) are not easily removed. One study by Friedman et al. 2020 examined the feasibility of stimulating microbial activity using hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) as a bio-specific and clean oxygen source that leaves no residuals in the water and is advantageous upon aeration due to the solubility limitations of the oxygen. The performance of a pilot bio-filtration system was enhanced by the addition of  $\text{H}_2\text{O}_2$  for particle, organic matter,  $\text{NH}_4^+$ , and  $\text{NO}_2^-$  removal. Hydrogen peroxide provided the oxygen demand for full nitrification. Results from Friedman et al. 2020 indicated that influent concentrations of  $4.2 \pm 2.5 \text{ mg/L N-NH}_4^+$  and  $0.65 \pm 0.4 \text{ mg/L N-NO}_2^-$  were removed during the short hydraulic residence time (HRT). In comparison, filtration without  $\text{H}_2\text{O}_2$  addition only removed up to  $0.6 \text{ mg/L N-NH}_4^+$  and almost no  $\text{N-NO}_2^-$ . A DNA metagenome analysis of the functional genes of the media biomass reflected a significant potential for simultaneous nitrification and denitrification activity. It is hypothesized that the low biodegradability of the organic carbon and  $\text{H}_2\text{O}_2$  addition stimulated oxygen utilization in favor of nitrification, followed by the enhancement of anoxic activity (Friedman et al. 2020).

### **PROJECT OBJECTIVE**

To reduce nitrogen concentrations, which would opportunistically starve harmful algal populations of nutrients that contribute to algal production and bloom formation, and increase dissolved oxygen concentrations in water ways around the City of Marco Island, Florida. Incidentally when harmful algal blooms produce toxins to the extent of being at or above safe recreational activities as defined by the World Health Organization (8 ppb), Lake Guard® Oxy's mode of action will also reduce toxin concentrations. The project's ultimate goal is to improve aquatic ecosystem health and consequently human and animal health and local economy.

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## TREATMENT AND SCOPE

### a) Material

Lake Guard® Oxy, a floating and time-releasing formulation of sodium percarbonate. The product slowly releases hydrogen peroxide which decomposes to release oxygen molecules and opportunistically induces oxidative stress to any harmful algae present in the water at the time of treatment. Use of Lake Guard Oxy will promote biodiversity in the aquatic ecosystem, as well as create a potential to stimulate submerged aquatic vegetation growth by also reducing nitrogen concentrations; collectively resulting in a healthier environment.

### b) Scope of Treatment

Proposed Geographic area: Internal waterways among the seven Marco Island Drainage Basin Areas, which equates to 1525 acres (Figure 4). In the spirit of this RFP, we aim to remediate an area of a significant size, thousands of acres, to maximize the benefits for the City of Marco Island and its stakeholders, and to demonstrate the scalable nature of BlueGreen’s technology. A treatment area of a scale of thousands of acres will utilize BlueGreen’s full suite of treatment and monitoring capabilities. Since water is a dynamic, rapidly changing system, the ideal way to develop an effective remediation effort, especially covering a large-scale area, is by acquiring high quality data about the conditions of the ecosystem, analyzing it effectively, formulating an action plan and executing it, while continuing to collect data to assess the quality of the treatment in near real-time, and devising further action items to act upon.

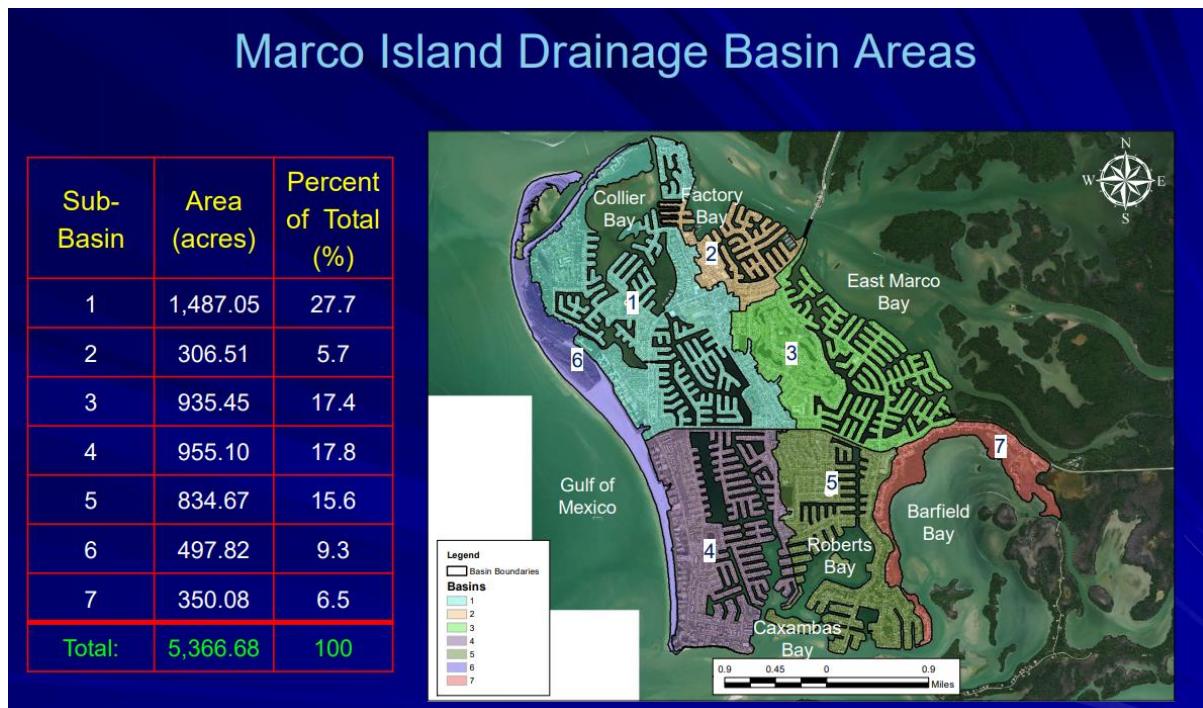


Figure 4. Marco Island Drainage Basin Areas (Credit: ERD Nutrient Source Evaluation Report 2023).

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### *c) Product Application*

Product application will be performed by licensed and insured applicators in the State of Florida. The ready to use Lake Guard® Oxy can be dispersed from the shores, a boat, or through areal application, at specified areas geographic areas that will allow the product to disperse, autonomously, with the movements of winds and currents. Lake Guard® Oxy application will follow the U.S. EPA-approved product label. BlueGreen scientific and technical team on the ground will advise on treatment regimen, localized areas for product dispersal, and on the frequency of treatment.

### *d) Permit Requirements*

The Lake Guard® Oxy is approved by the U.S. EPA, certified by the NSF/ANSI/CAN-60 for treatment in drinking water, and registered with the Florida Department of Agriculture and Consumer Services. It should be noted that BlueGreen is a Contractor (ES014) for the Florida Department of Environmental Protection (DEP) to provide Harmful Algal Bloom (HAB) Management Services on an as needed basis for its Districts. Nevertheless, BlueGreen will partner with local authorities to secure other local and state permits to apply the product, as needed. BlueGreen will follow the instructions of local authorities to the fullest degree.

## **MONITORING**

Monitoring is key in assessing the quality of treatment and to adapt the treatment, as needed, to optimize results. Monitoring will include the following parameters:

*a) Remote sensing* using satellite imagery and processing or aerial drone imagery that will be processed through BlueGreen's proprietary algorithm trained with real data using machine learning models. The remote sensing algorithms will help guide treatment regimen, treatment application sites, and treatment efficacy.

*b) In-situ field water measurements* with a handheld or automated multimeter:

- Water temperature
- pH
- Dissolved oxygen (DO)
- Chlorophyll-b (Chl-b, proxy for total biomass of green algae)
- Phycocyanin (PC, proxy for total biomass of cyanobacteria)
- Phycoerythrin (PE, proxy for total biomass of marine phytoplankton)
- Conductivity

*c) 3rd party laboratory work* to include Potentially Toxigenic (PTOX) screens and suite of water quality parameters.

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**PROPOSED COSTS AND COST STRUCTURE**

BLUEGREEN WATER TECHNOLOGIES, INC.					
Quote Number	JF040123a	<b>Water Body Name:</b>	Drainage Basin		
Quote Date	1-Apr-23	<b>Size in Acres:</b>	1525		
Agency Name	Marco Island, FL	<b>Period of Performance</b>	6 Months		
Proposal	RFI 2023-005				
Price Basis	<b>Fixed Pricing based on Scope and Scale</b>				
REMEDIATION SERVICE CONTRACT PRICING					
Item Code	Item Description	Unit	Unit Cost	Qty	Extended Price
Treatment	Prescribed Treatment Lake Guard Oxy® (30 days)	Per Acre	\$629.51	1525	\$960,000.00
Analytics	Comprehensive Analytics w/ Ground Truth (6 mo.)	Per Scope	\$290,000.00	1	\$290,000.00
Maintenance	Sustainment: Spot Treatment / Monitoring / Analytics	Monthly	\$249,750.00	5	\$1,248,750.00
<b>TOTAL - 6 Month Remediation including Monitoring Analytics</b>					<b>\$2,498,750.00</b>

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Product / Service	Component	In Scope	Quantity / Type	Provided By
Lake Guard View	Monitoring	BG Discretionary	1525 Acres	BlueGreen Option
Lake Guard Discover	Monitoring	Yes	7 Devices	BlueGreen
Lab Analysis	Monitoring	Yes	Up to 2x Monthly	BlueGreen
Drone Survey	Monitoring	Optional	As Needed	BlueGreen Option
Lake Guard Oxy	Treatment Product	Yes	As Needed	BlueGreen
Lake Guard Blue	Treatment Product	No	None	N/A
Lake Guard Dew	Treatment Product	No	None	N/A
Treatment Applicator	Field Services	Yes	As Needed	BlueGreen
Device Installation	Field Services	Yes	As Needed	BlueGreen
Device Maintenance	Field Services	Yes	As Needed	BlueGreen
Scientist	Consultation	Yes	10 Hrs / Mo	BlueGreen
Training	Professional Services	Optional	N/A	BG / Customer Option
Water Assessment	Professional Services	Yes	6 Months Analytics	BlueGreen
Science as a Service	Professional Services	Yes	Weekly Report	BlueGreen
Storage	Logistics	Yes	Warehouse	Customer
Jurisdictional Approvals	Project Management	Yes	As Needed	Customer
Community Outreach Campaign	Project Management	Yes	Media / Press	BlueGreen and Customer

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Harmful Cyanobacterial Bloom Mitigation in the C-43 Canal/ Caloosahatchee River Using BlueGreen Water Technologies (“BlueGreen”) *Lake Guard*<sup>®</sup> Oxy Solution

Amendments 2 & 3 to Agreement No. 35694 Between the SJRWMD and *BlueGreen* US for the Collaboration Between SJRWMD and SFWMD for the Treatment of Harmful Algal Blooms (HABs) in the C-43 Canal/ Caloosahatchee River

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Revised October 27, 2021

Lighter version, does not include the daily reports

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<b>Appendices E1-E15, daily reports, are not included in this lighter version.</b>	

# Harmful Cyanobacterial Bloom Mitigation in the C-43 Canal/ Caloosahatchee River Using BlueGreen Water Technologies (“BlueGreen”) *Lake Guard*<sup>®</sup> Oxy Solution

## *Remediation Project Report*

### **1. Abstract**

BlueGreen US Water Technologies (“BlueGreen”) was tasked by the South Florida Water Management District (“*SFWMD*”) to oversee and coordinate the mitigation of harmful algal blooms in a defined section of the C-43 Canal/ Caloosahatchee River. The mitigation protocols utilized the *Lake Guard*<sup>®</sup> Oxy, a unique formulation of sodium percarbonate-based algaecide that allows the active ingredient to float and time-release, to treat the cyanobacterial blooms. The *Lake Guard*<sup>®</sup> Oxy used in this project was obtained from the FL Department of Environmental Protection emergency stockpile maintained by *BlueGreen*. The main objective of the project was to mitigate cyanobacterial blooms in the residential canals around the C-43 Canal/ Caloosahatchee River. The residents of these canals had suffered from the adverse effects of algal blooms that include health risks, unpleasant sights, and a foul odor originating from floating rafts of decaying algae. The emergency applications provided an immediate relief for the residents, and the treatment eliminated the cyanobacterial scum and the associated odor, as well as other unpleasant conditions within 24-48 hours. Data indicates that the treatment application reduced significantly, to below threshold advisory levels established by the U.S. EPA of 8 µg/L (ppb) for recreational water, the cyanotoxin levels, which have reached high levels, well above threshold levels, in certain areas prior to treatment.

The initial project timeline was scheduled to be 7 days, overlapping with the Memorial Day weekend at the end of May, 2021, but due to the successful deployment and immediate results, the project was extended by another week, to a total of 15 days. The scope of treatments was extended to include all the residential areas along a 48-mile long segment of the C-43/ Caloosahatchee River, from Lake Okeechobee to Fort Myers. During the second week of treatments, *BlueGreen* introduced the *Lake Guard*<sup>®</sup> technology to *SFWMD* inspectors and *SFWMD* licensed contractor, and trained them on the application of the *Lake Guard*<sup>®</sup> Oxy.

Only 5.1 tons of the 20-ton stockpile were used over a 15-day period to remediate and maintain bloom free conditions in residential areas along the 48-mile river segment.

## **2. Background**

BlueGreen US Water Technologies (“*BlueGreen*”), together with the South Florida Water Management District (“*SFWMD*”) and Florida Department of Environmental Protection (“*FL DEP*”) have established a treatment regime for the remediation of the C-43 Canal/ Caloosahatchee River, in order to mitigate cyanobacterial blooms in specific sections of the canal, using *BlueGreen* Water Technologies *Lake Guard*<sup>®</sup> Oxy solution.

The C-43 Canal/ Caloosahatchee River is a mixed natural and manmade canal network, stretching from the banks of Lake Okeechobee, designated to lead water into the Atlantic Ocean and the Gulf of Mexico. When water in Lake Okeechobee reaches dangerous levels, the U.S. Army Corps of Engineers discharges water through various outlets to release pressure on the infrastructure. With water released, cyanobacterial blooms, prevalent in Lake Okeechobee, often proliferate into Florida’s waterways.

The C-43 canal (Caloosahatchee canal) along with the Caloosahatchee River extends 75 miles west of Lake Okeechobee, and is a major tributary to the Charlotte Harbor. It extends into the Gulf Coast, where it forms the Caloosahatchee Estuary.

The C-43 Canal includes 3 lock-and-dams; S-77 (Moore Haven Lock & Dam), immediately west of Lake Okeechobee’s rim canal, S-78 (Ortona Lock), 15.3 miles west of S-77, and S-79 (WP Franklin Lock and Dam), 27 miles west of S-78 (Fig. 1).

On May & June, 2021, the gates at S-77, S-78, and S-79 structures were open, and cyanobacteria-infested waters from Lake Okeechobee were flowing into the C-43 canal. This led to heavy blooms being introduced into the canal, in the vicinity of residential areas that suffered from the unpleasant sights and odor that accompanied the blooms.

## **3. Objective**

The initial objective and scope of this emergency project was to handle, contain and eliminate ongoing cyanobacterial blooms in the Caloosahatchee River segment downstream from the S-79 structure. The objective and scope of treatment were redefined on Thursday, May 27, during the *SFWMD* executive management visit, to focus on residential canals off of the Caloosahatchee River with visible cyanobacterial blooms and foul odor. The redefined objective was meant to immediately alleviate the residents’ predicament, and to preserve the *Lake Guard*<sup>®</sup> Oxy product for future emergency applications. The water salinity increases down the Caloosahatchee River, west of the S-79 structure, to the point of unfavorable conditions for cyanobacterial growth and survival. The scope of treatment expanded to beyond the residential canals west of the S-79 structure, to the residential areas east of the S-79 structure, as far as the S-77 structure.

#### 4. Executive Summary

This remediation project objective was to handle, contain, and eliminate ongoing cyanobacterial blooms in residential areas that span along a 48-mile length segment of the C-43 Canal/Caloosahatchee River, from the S-77 (Moore Haven Lock & Dam) to Fort Myers, in order to minimize their impact on health and quality of life

*BlueGreen's* technical experts together with Modica and Associates (*Modica*), licensed applicators, have assessed the C-43 Canal and its adjacent waterways in order to establish a proper mitigation protocol for the treatment regimen designed to target cyanobacterial outbreaks in progress, as well as to mitigate the developing conditions that may lead to such outbreaks in residential canals. Treatment was conducted with the *Lake Guard*<sup>®</sup> Oxy product, approved by the U.S. EPA, certified by the NSF/ANSI/CAN-60 for treatment in drinking water, and registered with the Florida Department of Agriculture and Consumer Services.



**Figure 1.** (A) A section of South Florida depicting Lake Okeechobee and the Gulf Coast. The area enclosed in the yellow box includes the 48-mile segment of C-43 Canal/ Caloosahatchee River area that was monitored and treated. (B) The 48-mile segment of the C-43 Canal/Caloosahatchee River where all residential areas were monitored and treated. The segment extends from the S-77 structure all the way to Fort Myers, downstream from the S-79 structure. Lock and Dam structures S-77, S-78, and S-79 are marked along the path of the canal. This image of the C-43 Canal/ Caloosahatchee River is courtesy of SFWMD GeoSpatial Services.

Over a 15-day period, *BlueGreen* inspected hundreds of residential canals and treated/ retreated a total of 56 sites along the 48-mile segment of the C-43 Canal/ Caloosahatchee River, resulting in fast bloom mitigation that provided immediate relief to the residents that suffered from heavy blooms near their

homes, and the associated foul odors and potential health risks. After the interventional treatments, and despite heavy bloom conditions in the C-43 Canal/ Caloosahatchee River, *BlueGreen* was able to maintain bloom free conditions in the residential areas along the C-43 Canal/ Caloosahatchee River with minimal dose rates.

Summary of the treatment applications over the 15-day project:

- Total cumulative Lake Guard Oxy applied: **10,260 lb (5.1 tons)**
- Total treatment and retreatment applications: **112**
- Total number of residential areas treated: **56**
- Covered length along the C-43/ Caloosahatchee River: **48 miles**

During this project, *BlueGreen* trained several inspectors from *SFWMD*, as well as a *SFWMD* subcontractor on treatment applications with the *Lake Guard*<sup>®</sup> Oxy.

This project demonstrated the efficacy and safety of the *Lake Guard*<sup>®</sup> technology in treating heavy cyanobacterial blooms and maintaining bloom free conditions in residential areas. The technology and its application could be deployed quickly, with immediate results. Staging for this operation took less than a day after the *SFWMD* secured a permit from the U.S. Army Corps of Engineers to stage the operation within its facilities and treat areas under their jurisdiction. Treatment results were evident within 24-48 post applications, namely, the removal of the cyanobacterial scum, and the elimination of the accompanied foul odor.

Notably, in certain areas, where cyanotoxin levels prior to treatment were well above the U.S. EPA advisory thresholds of 8 µg/L (ppb) for recreational water, the cyanotoxin levels were reduced significantly, to below advisory thresholds, after 24-48 hours from treatment.

Anecdotal evidence indicated that other unpleasant conditions in the residential areas were remediated as well beside the unwanted odor, e.g., the presence of flies that thrived on the decomposing floating scum.

*SFWMD* role in coordinating and facilitating the treatment applications with other Florida State agencies and the U.S. Army Corps of Engineers was critical in the immediate staging of this project and its ultimate success.

The efficacy and safety of the technology was demonstrated during heavy bloom conditions, however, *BlueGreen* recommends continuous preventative treatments to be conducted during early stages of evolving cyanobacterial outbreaks, which require lower doses of the active ingredient. The early

treatments are designed to eliminate cyanobacteria before the inevitable build-up of cyanotoxins during bloom episodes, thus securing a safe cleanup of the toxic species.

Treating the source, Lake Okeechobee, would prevent these cyanobacterial bloom episodes from recurring in Florida waterways. With proper planning, the *Lake Guard*<sup>®</sup> technology is scalable to treat HABs in Lake Okeechobee. As a first step, remediating a significant section of the Lake near the C-43 and C44 canals, will prevent contaminated waters from being released into these canals.

## 5. Staging

On Wednesday afternoon, May 26, 2021, 8 hours after *SFWMD* had secured a permit from the U.S. Army Corps of Engineers to allow *BlueGreen/ Modica* to stage an operational post at the W.P. Franklin South Recreation Area, *BlueGreen/ Modica* staging became operational and ready for monitoring and treatment applications. Monitoring, inspection, and treatment application started on Thursday, May 27, and continued daily through the Memorial Day weekend until Thursday June 10, 2021, a total of 15 days.

The initial amount of the *Lake Guard*<sup>®</sup> Oxy that was shipped to the operation site on Wednesday 26, 2021 was 9 tons, and two days later, another 18 tons of the *Lake Guard*<sup>®</sup> Oxy arrived at the operation site. The product was stored and secured in 4 containers.

*BlueGreen* and *Modica* field team consisted of 12 individuals, including 3 PhDs and 2 licensed applicators. *BlueGreen* also operated an offsite support team in Israel, of 5 individuals that included another 3 PhDs.

*Modica* secured a 24-hour staffing & monitoring of the staging area for 16 days.

Field operations started at 6:30 AM every day and lasted until after 8 PM, consisting of monitoring, inspection and treatment application over a 48-mile segment of the C-43 Canal/ Caloosahatchee River.

Over 2,300 hours were invested in this project, with approximately a third of the total hours (5/15 days) were holiday and weekend hours.

A total of 15 daily reports that detailed the operation and application treatments of each day were sent every night to *SFWMD*.

The staging site included the following:

- 27 tons of the *Lake Guard*<sup>®</sup> Oxy
- 4 x 20-ft containers
- A tent that served as headquarters
- Camper / “mobile staging area office”

- A Genie 1056 telehandler forklift
- A 31' Ameracat boat with tower, chart plotter with GPS tracker, 4kw radar, 1kw sonar w/ side scan, depth finder, water temperature sensor
- A pontoon boat with 400gal silo application setup
- Additional 1,100 gal silo
- 3 x trucks
- 4 x SUVs



**Figure 2.** Staging at the W. P. Franklin South Recreational Area. Wednesday, May 26 – Friday June 11, 2021. As part of the great effort to mitigate the blooms occurring throughout the C-43 Canal in a fast and efficient manner, *BlueGreen/ Modica* have conducted an extensive remediation project, in which 56 different sites were sampled and treated with 112 separate treatments and retreatments, using a total amount of 10,260 lb (5.1 tons) of the *Lake Guard*® Oxy product. In order to achieve this highly ambitious goal within only two weeks, *BlueGreen/ Modica* labored around the clock. A campsite and a command post were established in the field in order to assist with the extensive sample collection, the logistical issues, and handing the equipment.





**Figure 3.** Treatment applications in residential areas along a 48-mile segment of the C-43/ Caloosahatchee River. Thursday, May 27 – Thursday, June 10, 2021.

## 6. Treatment

The *Lake Guard*<sup>®</sup> Oxy product is a targeted treatment against cyanobacteria, selectively eliminating and preventing toxic algal blooms by triggering a biological programmed cell death within the cyanobacterial communities, resulting in a population-wide collapse of the cyanobacteria. The product is a granular material composed of 98% (w/w) sodium percarbonate that releases hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), and 2% (w/w) of an inert, biodegradable encapsulating agent that floats and time-releases the active ingredient on the water surface, homing-in on cyanobacterial aggregates as they drift in the water. The product can be applied manually from the shore, a boat, or a plane, and is scalable to large water bodies.

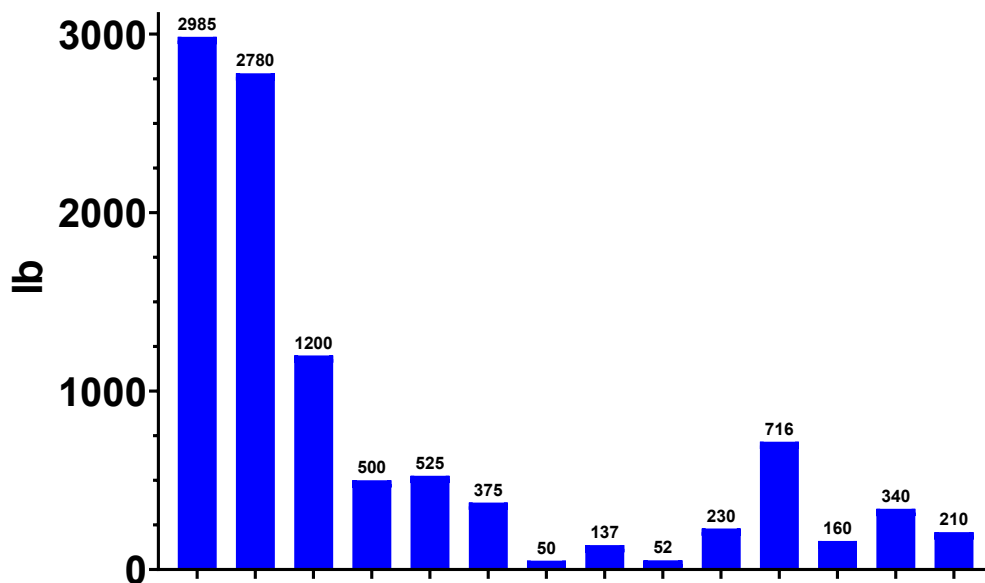
In general, the application rates for the *Lake Guard*<sup>®</sup> Oxy range between 0.5-30 lb/acre, depending on the intensity of the bloom and other conditions in the water. The lower doses are reserved for preventative treatments, for example, the application rates in Lake Minneola (1,890 acres) ranged between 0.5-7.5 lb/acre, with an average rate of approximately 1 lb/acre.

Most of the residential areas around the C-43/ Caloosahatchee River were heavily contaminated with cyanobacterial scum, with an estimated cyanobacterial cell count between 10<sup>6</sup>-10<sup>9</sup> cells/ml, often times at

the upper limit of that range. Due to the urgent nature of this project that overlapped with the Memorial Day weekend, and the limited time available to remediate residential canals over a river segment of 48 miles, *BlueGreen* increased its usual dosage rates to ensure immediate relief for the residents. The average dosage rate over the 15-day project was about 40 lb/acre, with a range of 0.8 – 181.8 lb/acre. The larger dose rates were not anywhere near the maximum dosage rate of 294 lb/acre approved by the label. Due to heavy boating activity on weekends, the Army Corps of Engineers permit limited the treatment applications between 6:30-8:30 AM, and between 5-8 PM during the Memorial Day Weekend, and on the weekend that followed.

The total amount of the Lake Guard® Oxy product used for the full course of the remediation project was 10,260 lb (Fig. 4), applied over 112 different treatments and retreatments, at 56 different residential sites. The amount of product required for maintaining bloom free conditions in the residential canals declined substantially after the initial interventional treatments (Fig. 4).

## Lake Guard® Oxy use for daily treatments



**Figure 4.** Daily amounts of *Lake Guard*® Oxy product used in the course of the 15-day treatment application, along the 48-mile segment of the C-43/ Caloosahatchee River. Most of the product was used during the interventional period, at the early stages of the project. Maintaining bloom-free canals after the interventional treatment and expanding to new areas required lower application rates.

## 7. Sampling

Monitoring is key to follow bloom progression, assess the quality of the treatment, and adapt the treatment as needed to optimize its results. Due to the nature of this project, most of the monitoring consisted of visual inspection of the residential canals, but additional extensive monitoring took place in

selected areas, which included the following parameters: in-situ field water measurements with a YSI ProDSS multiparameter; Sampling of water temperature, pH, dissolved oxygen (DO), chlorophyll-b (Chl-b; proxy to the total biomass of green algae), phycocyanin (PC, proxy to the total biomass of cyanobacteria), and conductivity; Field microscopy of samples obtained from the water surface, for detection of the variety of cyanobacterial species present in the water; Laboratory work to include taxa analysis and presence of toxins.

## ***8. Results***

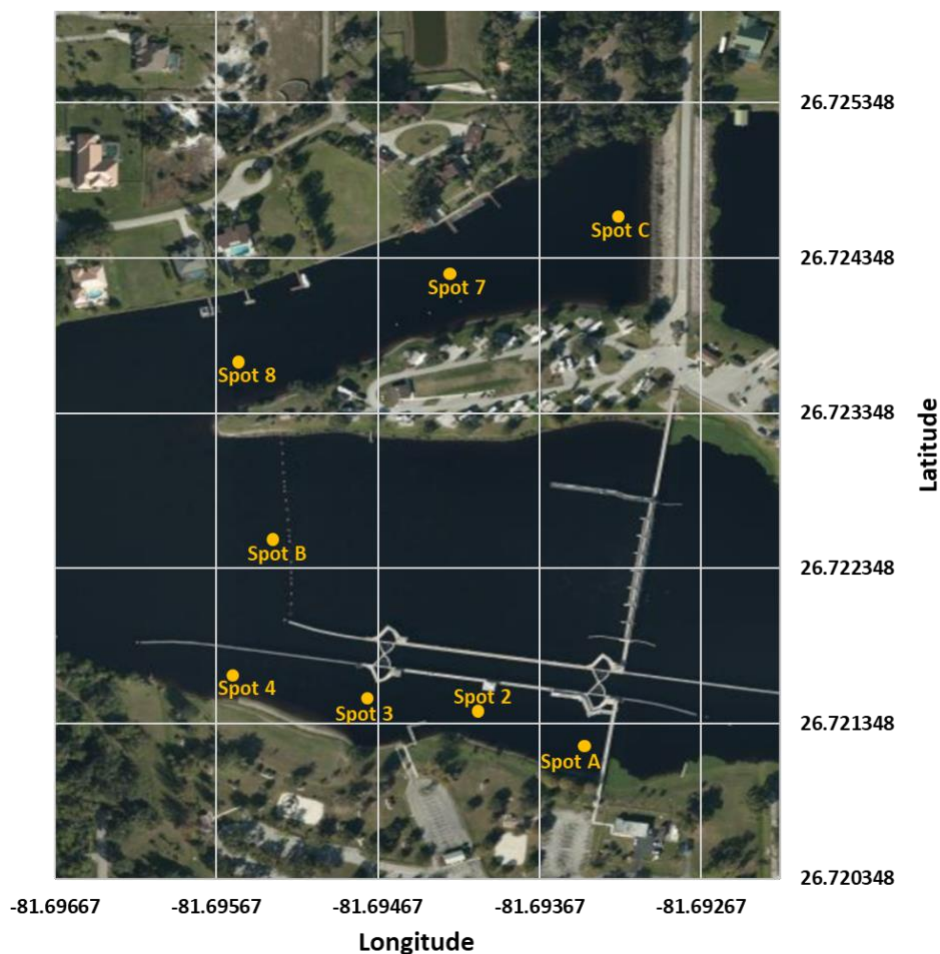
In total, *BlueGreen/ Modica* inspected, treated and maintained residential canals over a 48-mile long river segment, conducting a total amount of 112 treatments/ retreatments at 56 different sites.

The treatments provided immediate relief to the residents that suffered from heavy blooms near their homes and a strong foul odor that accompanied the blooms. Total application rates, maps, and pictures from each of the 56 sites are detailed in Appendix A.

Results of the daily measurements of toxins in samples A, B, C, 2, 3, 4, 7, and 8, located around the W.P. Franklin Lock & Dam (Fig. 5), are presented in Table 1. While saxitoxins values exhibit no clear trend, a general trend of decrease in toxin levels is demonstrated for microcystins / nodularins. This trend is also observed in Fig. 6, where microcystins / nodularins in sampling spots A, B, and C exhibit a clear decline following treatment with the *Lake Guard*<sup>®</sup> Oxy product.

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### W.P. Franklin Lock and Dam (S-79)



**Figure 5.** Map of the W.P. Franklin Lock and Dam (S-79) and the sampling points along the S-79 structure. Spots A, B, C, 2, 3, 4, 7, and 8 were chosen as daily sampling points for in situ field water measurements, microscopy, visual inspections, and lab measurements of algal taxa and toxins. Potentially Toxicogenic (PTOX) cyanobacteria species *Aphanizomenon/ Chrysochlorum / Sphaerospermopsis sp.*, *Cuspidothrix sp.*, *Dolichospermum sp.*, *Microcystis sp.*, *Planktothrix sp.*, *Pseudanabaena sp.*, and *Raphidiopsis sp.*, detected in sampling spots A, B, C, 2, 3, 4, 7, and 8, exhibited a substantial decrease, following treatment with the *Lake Guard® Oxy* (Fig. 7).

**Table 1.** The cyanobacteria- associated toxins, microcystins / nodularins and saxitoxins concentrations (ng/mL), found in samples A, B, C, 2, 3, 4, 7, and 8, located at the S-79 structure, during May 2021.

Spot A	microcystins / nodularins	saxitoxins	Spot 2	microcystins / nodularins	saxitoxins
28 May	8.2	0.08	28 May	2.33	0.11
29 May	7.94	0.09	29 May	3.59	0.07
30 May	0.27	0.1	30 May	0.8	0.1
31 May	0.48	0.11	31 May	2.02	0.1

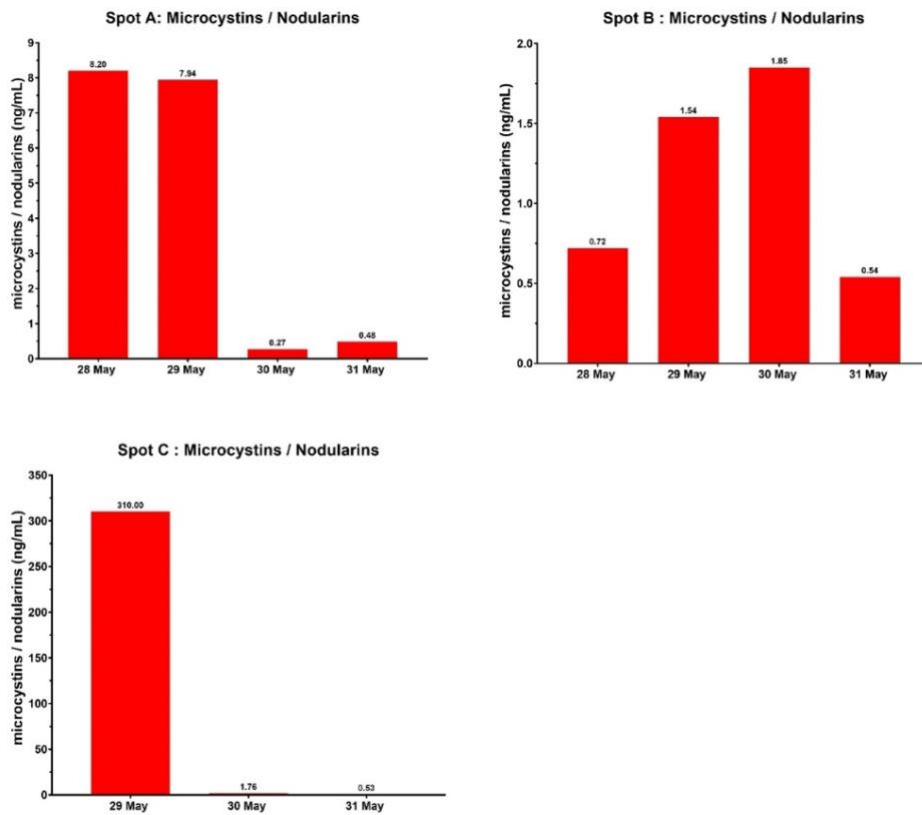
<b>Spot 3</b>	microcystins / nodularins	saxitoxins	<b>Spot 4</b>	microcystins / nodularins	saxitoxins
28 May	2.44	0.09	28 May	0.9	0.09
29 May	0.99	0.09	29 May	1.01	0.1
30 May	0.8	0.1	30 May	2.91	0.1
31 May	0.99	0.09	31 May	0.7	0.1

<b>Spot B</b>	microcystins / nodularins	saxitoxins	<b>Spot C</b>	microcystins / nodularins	saxitoxins
28 May	0.72-+0.2	0.09	28 May	---	---
29 May	1.54	0.1	29 May	310	0.07
30 May	1.85	0.11	30 May	1.76	0.09
31 May	0.54	0.09	31 May	0.53	0.1

<b>Spot 7</b>	microcystins / nodularins	saxitoxins	<b>Spot 8</b>	microcystins / nodularins	saxitoxins
28 May	---	---	28 May	---	---
29 May	1.55	0.09	29 May	1.46	0.09
30 May	0.7	0.08	30 May	0.73	0.08
31 May	0.67	0.1	31 May	4.08	0.08

Images of selected species of Potentially Toxigenic Cyanobacteria found in samples collected at spots 1, 4, and 7 are presented in Fig. 8.

Fig. 9 summarizes all in-situ digital measurements taken around the W.P. Franklin Lock & Dam. Water temperatures fluctuated around ~28°C (Fig. 9A); Conductivity levels gradually increased from ~870 µS/cm to ~1100 µS/cm in most sampling spots, however, some remained unchanged (Fig. 9B). Dissolved oxygen levels remained steady during the sampling periods in all spots, at values between ~80% and 100% (Fig. 9C). pH levels varied between 8 and 8.5 (Fig. 9D). Chlorophyll levels were maintained at ~10- 20 µg/L (Fig. 9E). Phycocyanin (PC) levels averaged around 3 µg/L (Fig. 9F).



**Figure 6.** Microcystins / nodularins found in samples A, B, and C, located at the S-79 structure (Fig. 5), during May 2021.

Appendix B includes all the lab reports for samples collected around the W.P Franklin Lock & Dam.

The reports include PTOX screen and cyanotoxin levels.

The name and ID of few of the water samples in this report were changed for simplicity reasons, and to avoid confusion from the ID that appears in the lab report (Appendix B). Below are the changes to the naming of these samples.

Sample 1 in the lab report, Appendix B, was renamed to Spot A in this report.

Sample 2 in the lab report, Appendix B, remained as Spot 2.

Sample 3 in the lab report, Appendix B, remained as Spot 3.

Sample 4 in the lab report, Appendix B, remained as Spot 4.

Sample 5 in the lab report, Appendix B, was renamed to Spot B in this report.

Sample 6 in the lab report, Appendix B, was renamed to Spot C in this report.

Sample 7 in the lab report, Appendix B, remained as Spot 7.

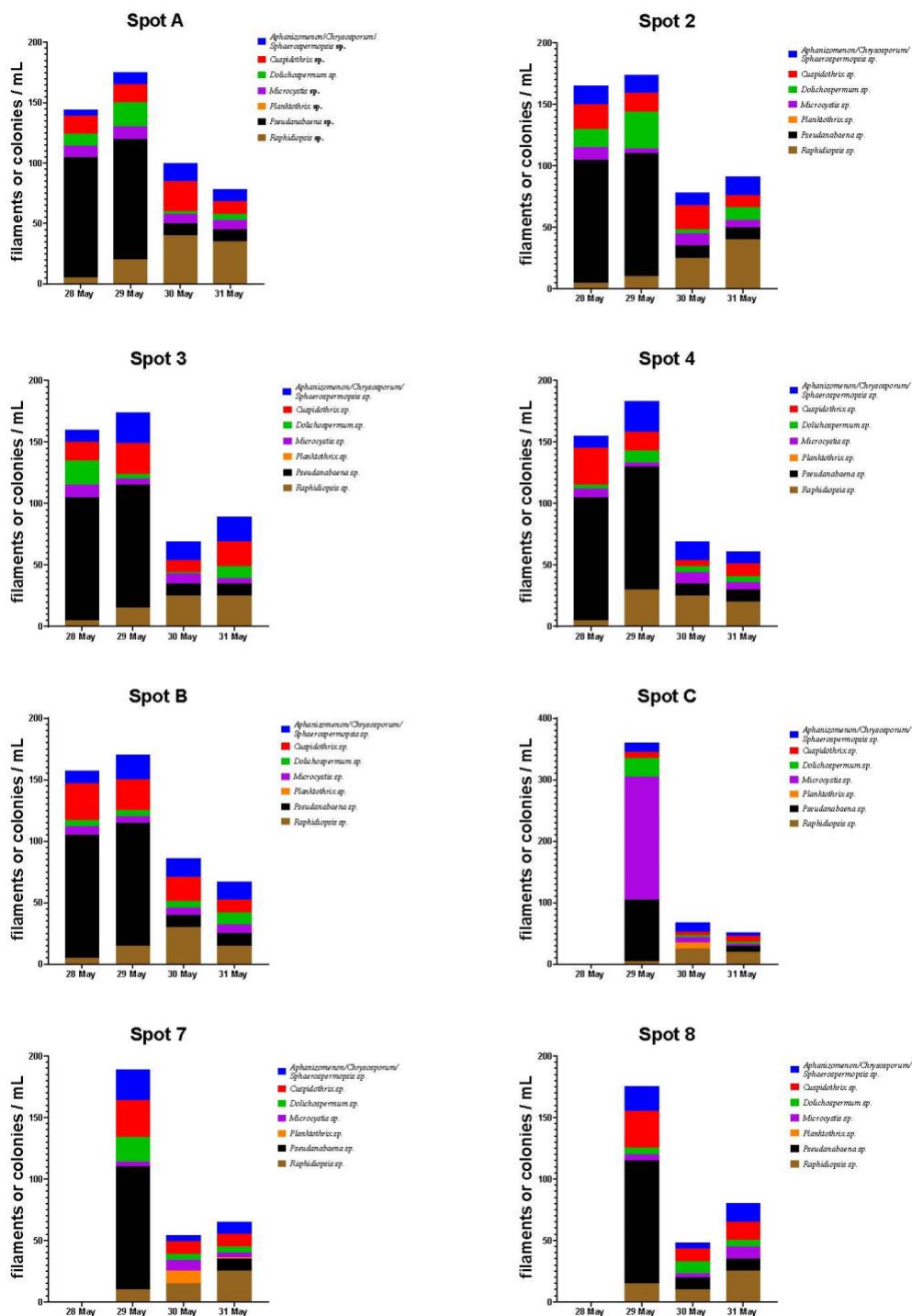
Sample 8 in the lab report, Appendix B, remained as Spot 8.

Sample 9 in the lab report, Appendix B, is the water sample from the treatment site #1, the farthest point to the west, Table 2.

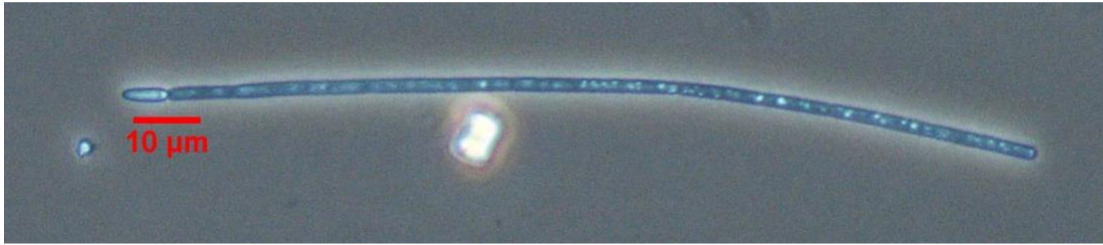
Sample 10 in the lab report, Appendix B, is a sample point on the east side of the W.P. Franklin Lock & Dam, point 33 (Fig.) 11.

Sample 12 in the lab report, Appendix B, is a sample point on the east side of the W.P. Franklin Lock & Dam, point 33 (Fig.) 11.

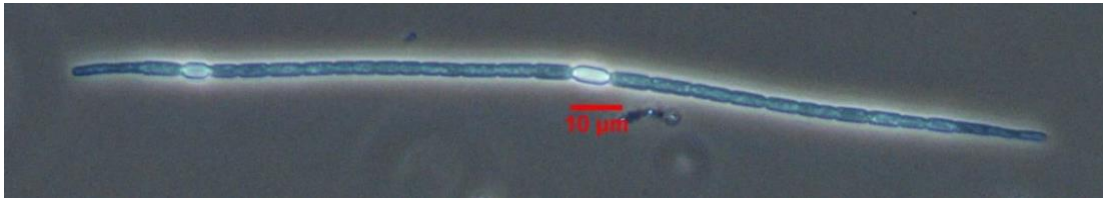
## Potentially Toxicogenic (PTOX) Cyanobacteria



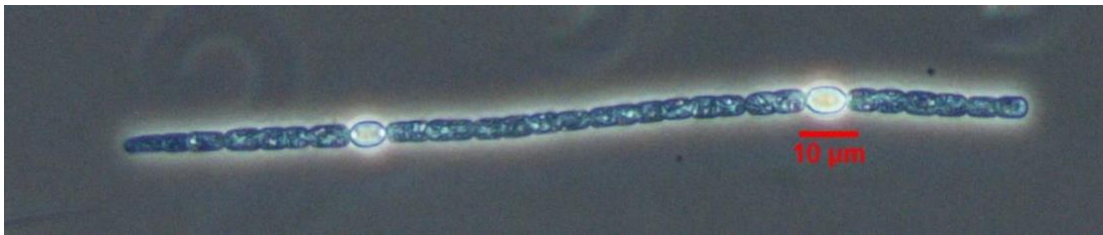
**Figure 7.** Potentially Toxicogenic (PTOX) cyanobacteria filaments/ colonies (per mL), found in samples A, B, C, 2, 3, 4, 7, and 8, during May 2021. The following species were detected: *Aphaniizomenon/Chrysosporum/Sphaerospermopsis sp.*, *Cuspidothrix sp.*, *Dolichospermum sp.*, *Microcystis sp.*, *Planktothrix sp.*, *Pseudanabaena sp.*, and *Raphidiopsis sp.*



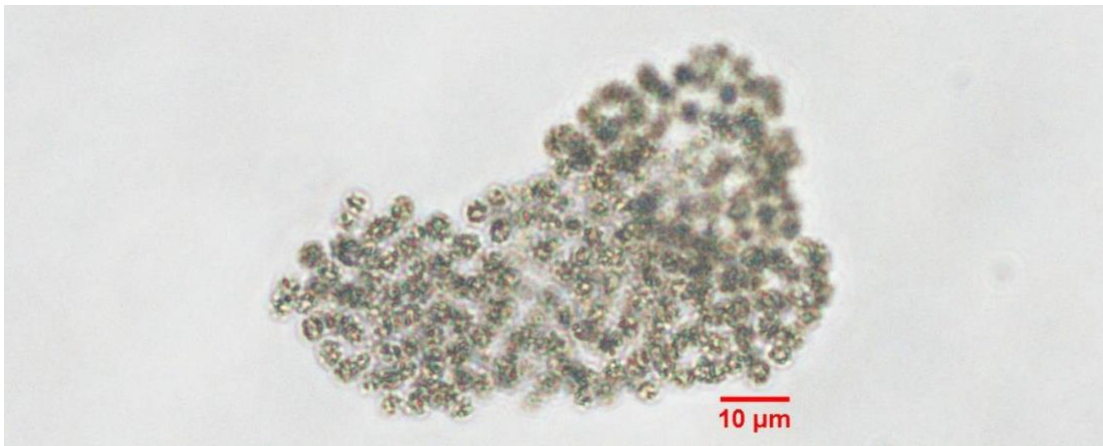
*Raphidiopsis* sp. (straight) at 400X (Spot 1)



*Cuspidothrix* sp. at 400X (Spot 1)

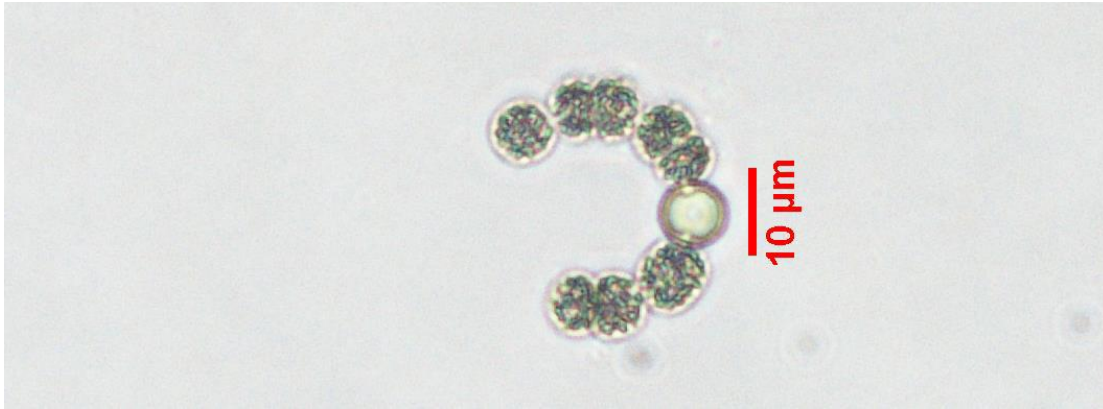


*Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. at 400X (Spot 1)



*Microcystis* sp. at 400X (Spot 1)

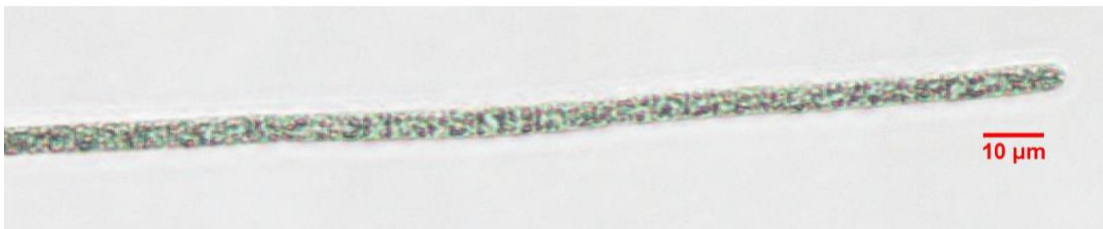




*Dolichospermum* sp. at 400X (Spot 1)

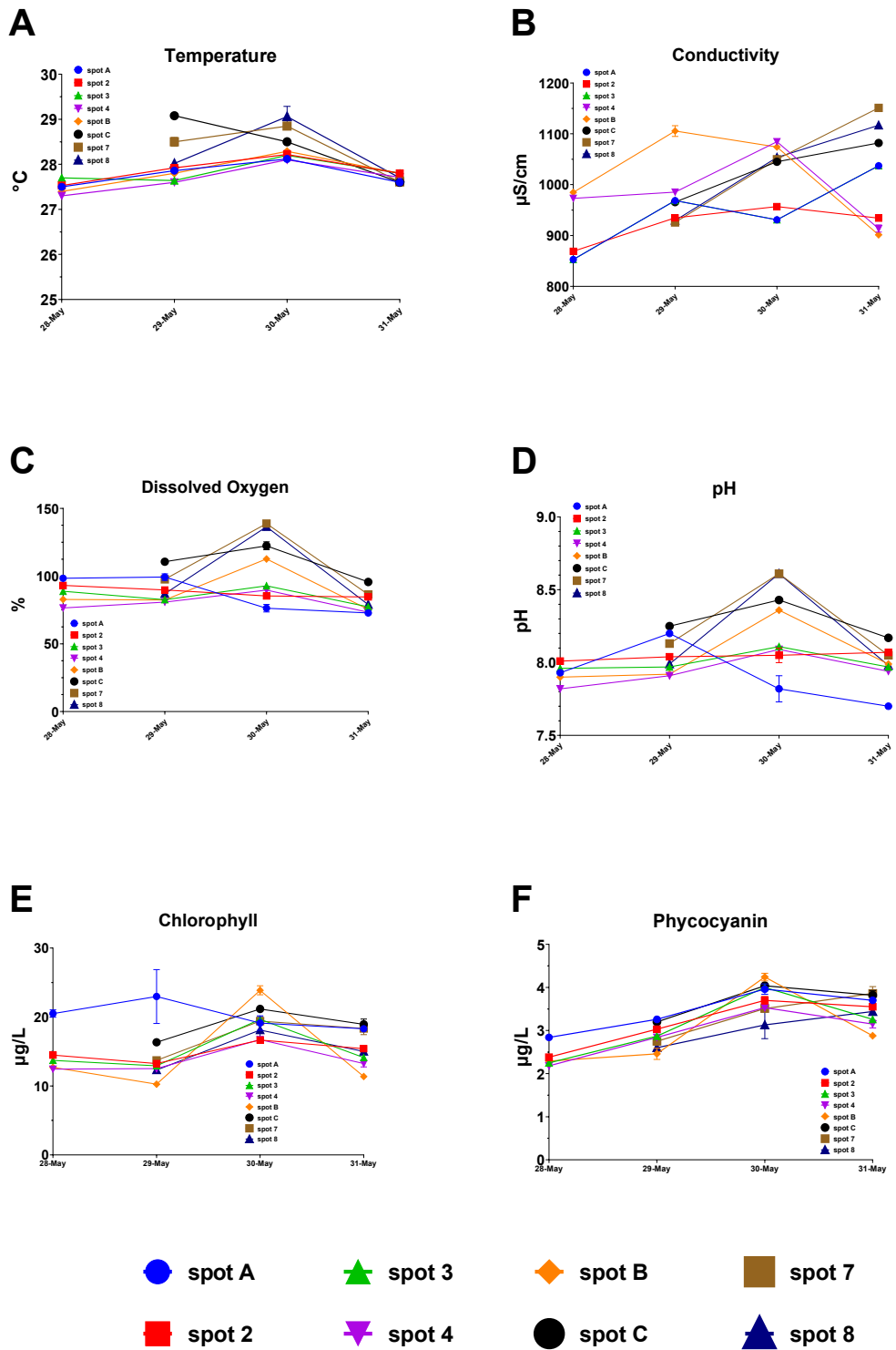


*Pseudanabaena* sp. at 400X (Spot 4)



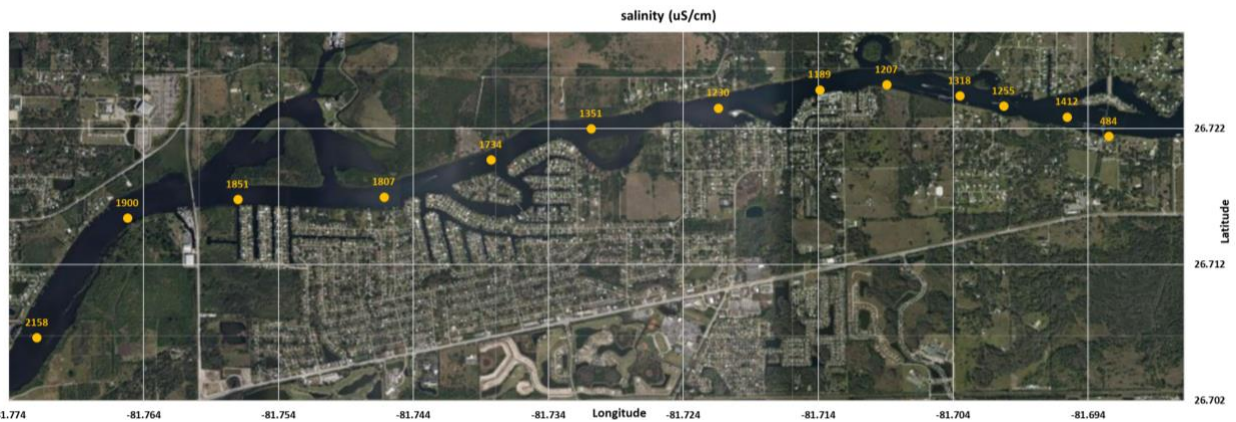
*Planktothrix* sp. at 400X (Spot 7)

**Fig. 8.** Selected micrographs of various species of Potentially Toxic Cyanobacteria (PTOX) found in samples collected at spots 1, 4, and 7.



**Figure 9.** In-situ field measurements obtained with the YSI ProDSS multimeter during May 2021. (A) water temperature (°C), (B) conductivity (µS/cm), (C) dissolved oxygen (%), (D) pH, (E) chlorophyll (µg/L), and (F) phycocyanin levels (µg/L, as a proxy for total cyanobacteria).

Measurements west of the S-79 lock and dam showed that the conductivity values increase along the Caloosahatchee river, west of the S-79 structure, with a gradient across the S-79 lock & Dam (Fig. 10; Table 2)



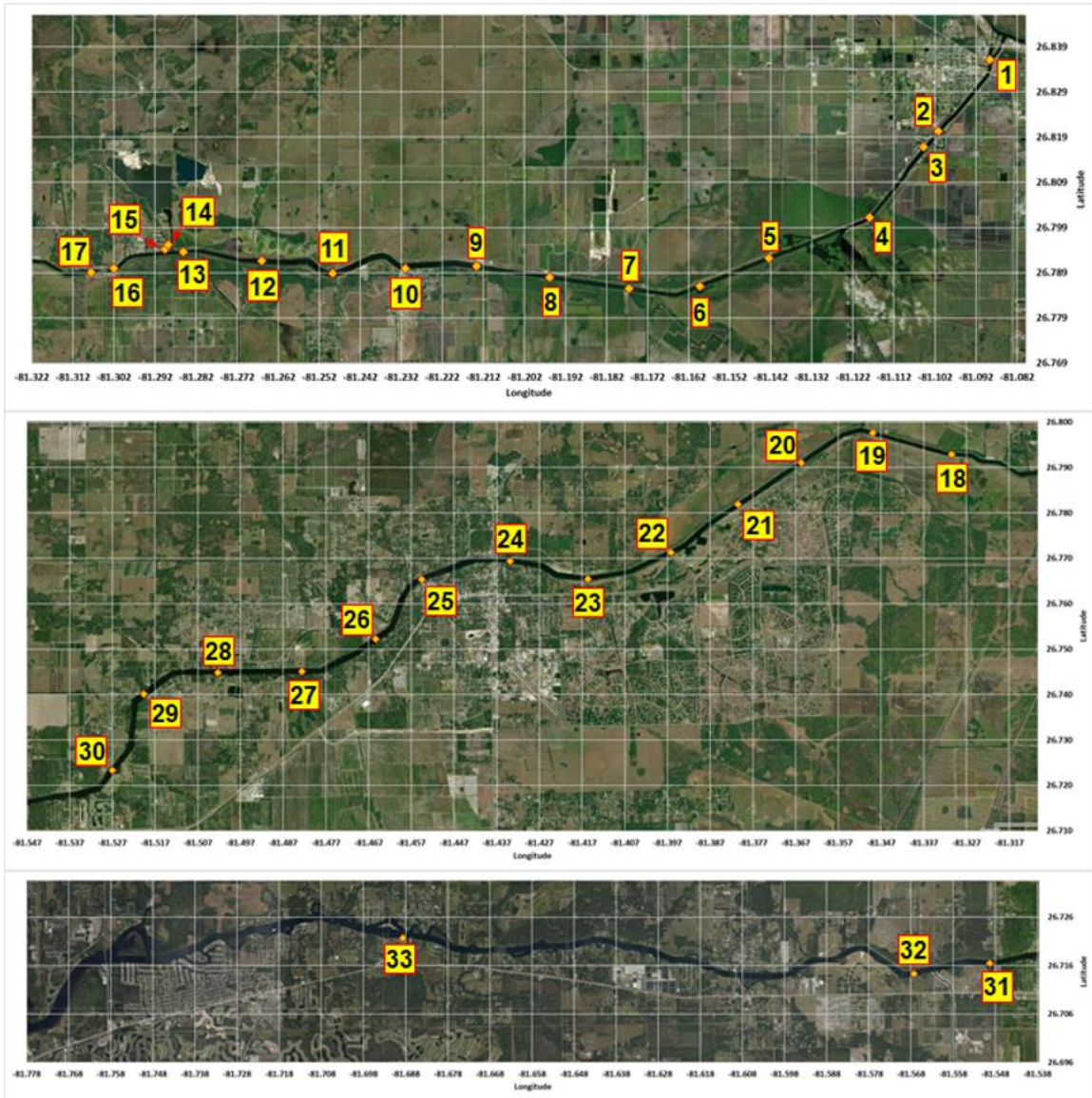
**Figure 10.** Data points across a 5.5-mile segment on the Caloosahatchee River, from the S-79 structure and downstream, where YSI ProDSS measurements were taken on May 28, 2021. Numbers indicate conductivity in  $\mu\text{S}/\text{cm}$ . Conductivity increases along the path downstream from the S-79 structure, with a gradient across the S-79 structure; 484  $\mu\text{S}/\text{cm}$  on the east side, and 1412  $\mu\text{S}/\text{cm}$  on the west side.

**Table 2:** Water parameter values on the Caloosahatchee River, from the S-79 structure to Fort Myers, a 5.5 mile segment. YSI-ProDSS field measurements from sampling points indicated in Fig. 10, May 28, 2021. Point 1 is the farthest point to the west. Point 13 is located on the east side of the S-79 structure. Temperature ( $^{\circ}\text{C}$ ), dissolved oxygen (DO %), conductivity (C- $\mu\text{S}/\text{cm}$ ), pH, chlorophyll (Chl  $\mu\text{g}/\text{L}$ ), phycocyanin (PC  $\mu\text{g}/\text{L}$ ), and latitude and longitude are presented. Note, the numbering refers to Fig. 10, and is different sample points than those in Fig. 5.

Point	$^{\circ}\text{C}$	DO %	C- $\mu\text{S}/\text{cm}$	pH	Chl $\mu\text{g}/\text{L}$	PC $\mu\text{g}/\text{L}$	Lat	Lon
1	29.30	139%	2158	8.41	14.72	1.42	26.70633	-81.77222
2	29.90	149%	1900	8.50	14.07	1.53	26.71513	-81.76551
3	29.40	142%	1851	8.46	16.92	2.45	26.71649	-81.75734
4	30.05	162%	1807	8.63	17.75	2.30	26.71667	-81.74649
5	30.30	180%	1734	8.76	24.82	3.13	26.71939	-81.73857
6	29.90	156%	1351	8.63	20.04	2.82	26.72169	-81.73114
7	29.90	158%	1230	8.64	18.93	3.10	26.72320	-81.72172
8	29.81	150%	1189	8.58	17.88	2.95	26.72454	-81.71420
9	29.80	149%	1207	8.56	17.45	3.23	26.72493	-81.70924
10	29.38	133%	1318	8.44	16.90	3.06	26.72411	-81.70382
11	29.41	141%	1255	8.49	18.19	3.22	26.72336	-81.70057
12	28.85	119%	1412	8.30	17.08	3.03	26.72255	-81.69586
13	30.30	139%	484	8.58	16.66	2.79	26.72112	-81.69278

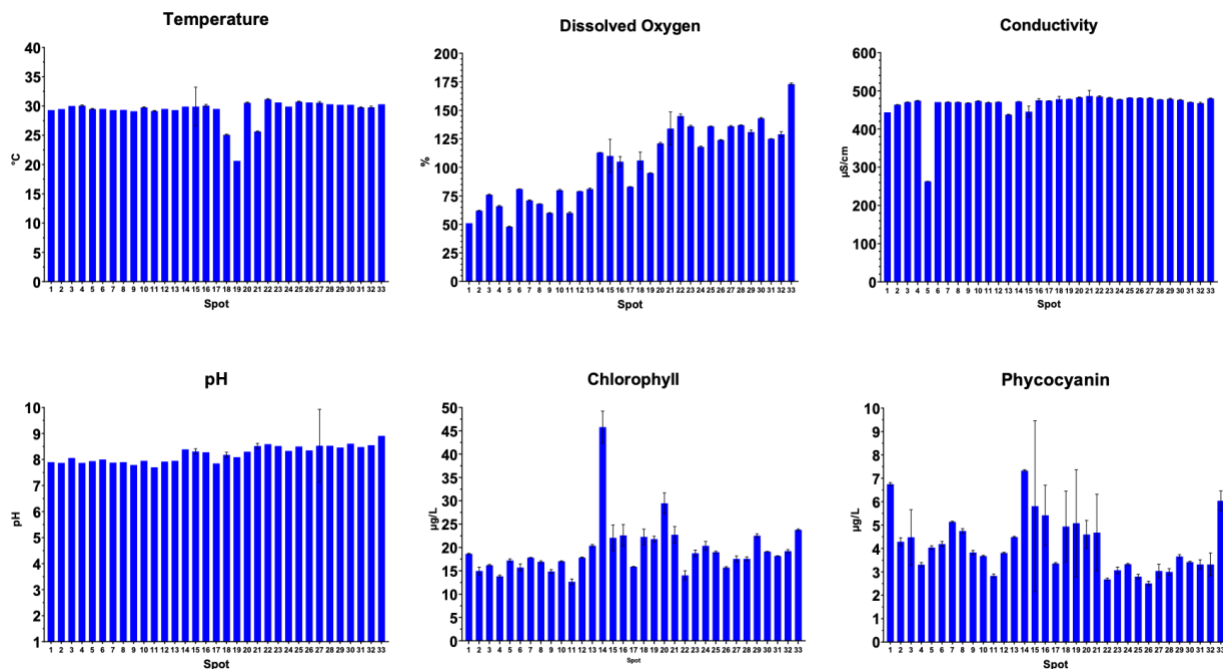
Since the Army Corps of Engineers permit limited the treatment applications to between 6:30-8:30 AM, and between 5-8 PM during the Memorial Day Weekend, and on the weekend that followed, *BlueGreen/Modica* utilized the time between treatments on weekend for extensive monitoring.

On Sunday, June 6, *BlueGreen/Modica* conducted a widespread inspection of all the residential canals east of the S-79 lock & dam, and collected extensive data along the C-43/ Caloosahatchee River from the S-77 lock & dam to the S-79 lock and dam (Fig. 11). Stormy weather interfered with data collection.



**Figure 11.** YSI-ProDSS sampling points along the C-43/ Caloosahatchee River, from S-77 structure (sample 1), to the S-79 structure (sample 33). Stormy weather interfered with data collection. Laboratory samples for PTOX screen and cyanotoxin analyses were collected at sampling points 1, 7, 16, 17, 20, 24, and 28

Water temperature, conductivity, and pH remained almost stable across the length of the river. Dissolved oxygen levels increased along the river, from east to west. Chlorophyll and phycocyanin levels fluctuated across the length of the river.



**Figure 12.** YSI-ProDSS field measurements for points 1 through 33, from the S-77 to the S-79 structure. Water temperature (°C), conductivity (µS/cm), dissolved oxygen (%), pH, chlorophyll (µg/L), and phycocyanin levels (µg/L, as a proxy for total cyanobacteria) were monitored.

Laboratory samples were collected from 7 sampling points across the length of the canal for PTOX screen and cyanotoxin analysis. The samples are 1, 7, 16, 17, 20, 24, and 28 (Fig. 11).

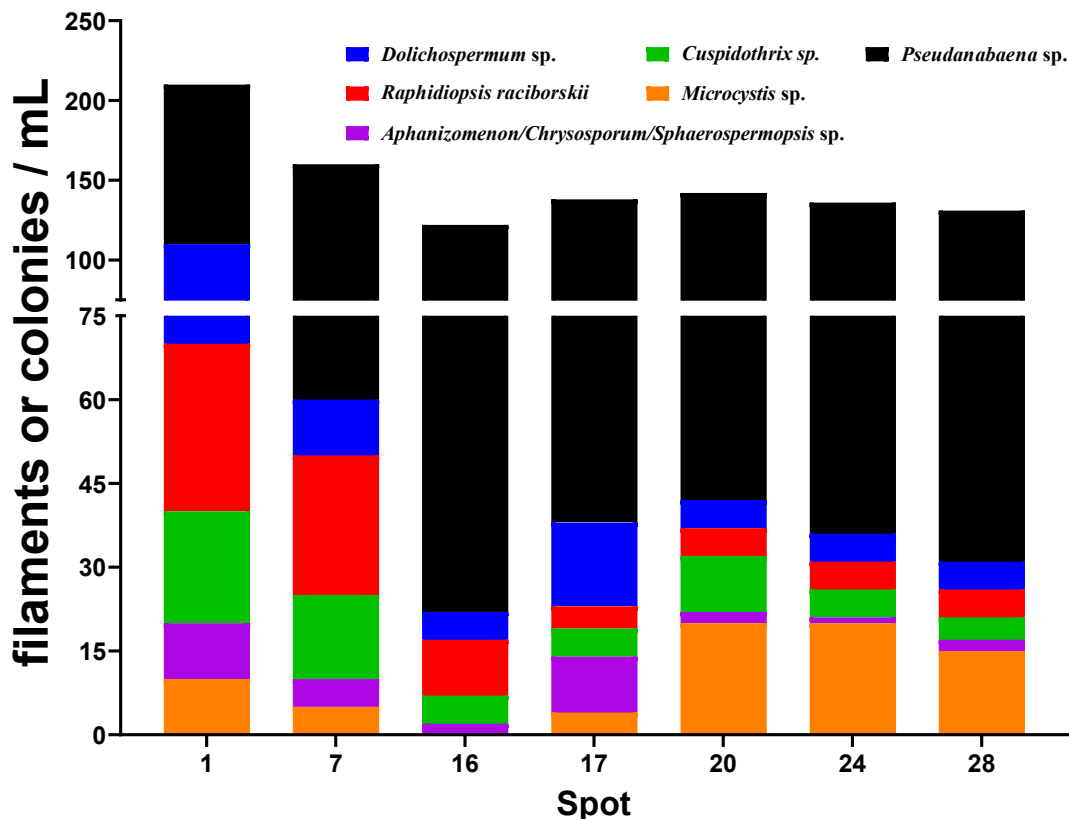
The PTOX screen results are summarized in Fig. 13. Sample 16, on the east side of S-78 exhibited extremely high levels of microcystins/ nodularins (Fig. 14). Samples 1 and 17 had microcystins/ nodularins levels above the U.S. EPA advisory threshold of 8 µg/L (ppb) for recreational water.

Lab reports from samples 1, 7, 16, 17, 20, 24, and 28, are appended to this report as appendix C.

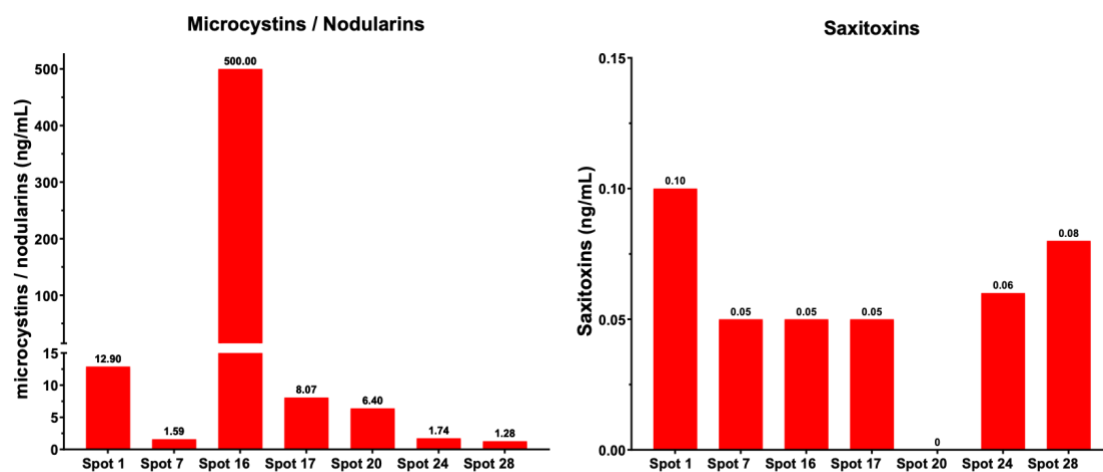
The name and ID of the water samples in this report were changed for simplicity reasons, and to avoid confusion from the ID that appears in the lab report (Appendix C). Below are the changes to the naming of these samples.

Samples 31 in the lab report, Appendix C, was renamed to Sample 1 in this section.  
 Samples 32 in the lab report, Appendix C, was renamed to Sample 7 in this section.  
 Samples 33 in the lab report, Appendix C, was renamed to Sample 16 in this section.  
 Samples 34 in the lab report, Appendix C, was renamed to Sample 17 in this section.  
 Samples 35 in the lab report, Appendix C, was renamed to Sample 20 in this section.  
 Samples 36 in the lab report, Appendix C, was renamed to Sample 24 in this section.  
 Samples 37 in the lab report, Appendix C, was renamed to Sample 28 in this section.

# PTOX



**Figure 13.** PTOX screen summary for water samples collected along the C-43/ Caloosahatchee River, from the S-77 to the S-79 structure (Fig. 11).



**Figure 14.** Cyanotoxin analyses of water samples collected along the C-43/ Caloosahatchee River, from the S-77 to the S-79 structure (Fig. 11).

The weather throughout the duration of the project was mostly clear to partly cloudy skies, with few shower storms, as indicated in the 15 daily reports (Appendices E1-E15). *BlueGreen* measured the flow of the river waters in several instances, and it had appeared to change from one measurement to another. However, these measurements were unreliable, because they were taken from the boat's surface, which was difficult to maintain at a fixed position. Flushing at the deep end of the residential canals was unlikely, although river waters flowing into the entry point of the canal were observed to have different rates, depending on the wind, currents, and the angle of the entry point.

## ***9. Case study – site 43***

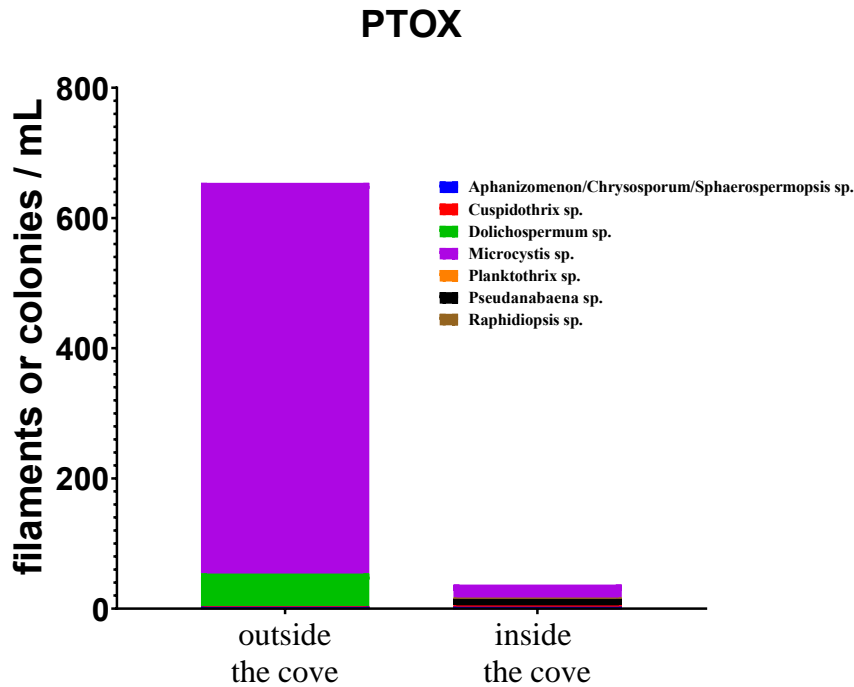
Out of the 56 sites that were treated during the 15-day intensive campaign, several sites were subject to thorough investigation. Such is the case of sites 38 and 43 (see details of location, images and analysis of site 38 and 43 below, in Appendix A: An overview of the 56 individual sites treated by BlueGreen/Modica).

Site 43 included is a cove that was first treated on Friday, June 4, 2021. Water samples were obtained from outside and inside the cove for extensive comparison of PTOX values, genome sequencing, and cyanotoxin analyses. The samples were collected two days after a treatment application inside the cove. The treatment had cleared the cyanobacterial scum, and no visible cyanobacterial aggregated were detected in the waters inside the cove one day after treatment. The data indicates that the treatment had significantly decreased cyanobacterial biomass and increased the biodiversity inside the cove compared to outside the cove, at the Caloosahatchee River.

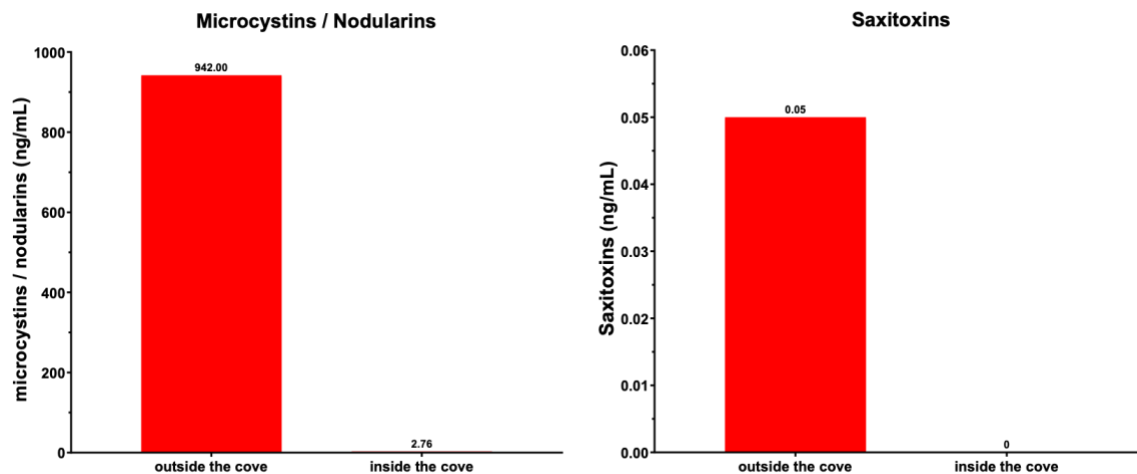
Comparison between PTOX values inside and outside the cove shows a substantial difference between PTOX species. It is evident that PTOX filaments / colonies counts are significantly more abundant outside the cove (Fig. 15). A similar pattern is observed for the toxins measured in this study, i.e., microcystins / nodularins and saxitoxins, which both exhibit sustainably lower values inside compared to outside the cove (Fig. 16, Appendix B, note the corresponding samples from the lab report below).

- Sample K20 in the lab report, Appendix B, is the water sample from outside the cove at the treatment site #43.
- Sample K21 in the lab report, Appendix B, is the water sample from inside the cove at the treatment site #43.

YSI-ProDSS field measurements are summarized in Fig. 17. The decrease in phycocyanin and the increase in chlorophyll is consistent with other results, and with the visual inspection of decreased cyanobacterial load and the increase in biodiversity.

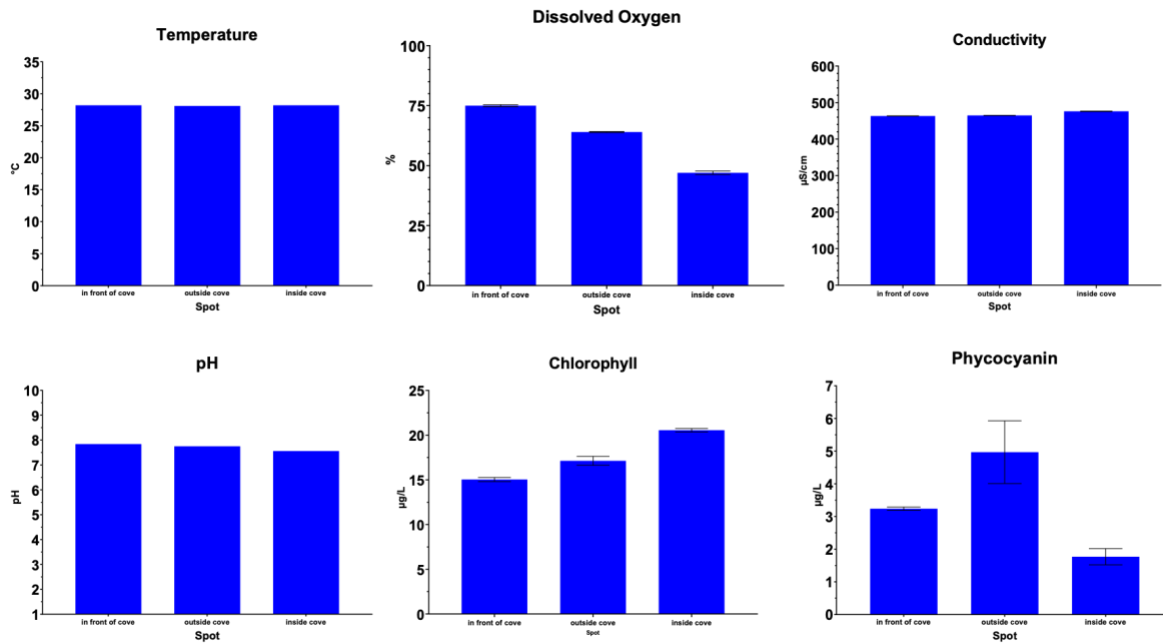


**Figure 15.** Potentially Toxicogenic (PTOX) cyanobacteria filaments/ colonies (per mL), found inside and outside the cove in site 43 on May 2021. Comparison between the following species was conducted: *Aphanizomenon/ Chrysochlorum / Sphaerospermopsis sp.*, *Cuspidothrix sp.*, *Dolichospermum sp.*, *Microcystis sp.*, *Planktothrix sp.*, *Pseudanabaena sp.*, and *Raphidiopsis sp.*



**Figure 16.** Microcystins / nodularins (top panel) and saxitoxins (bottom panel) (in ng/mL) found in samples taken inside and outside the cove on site 43, during May 2021.



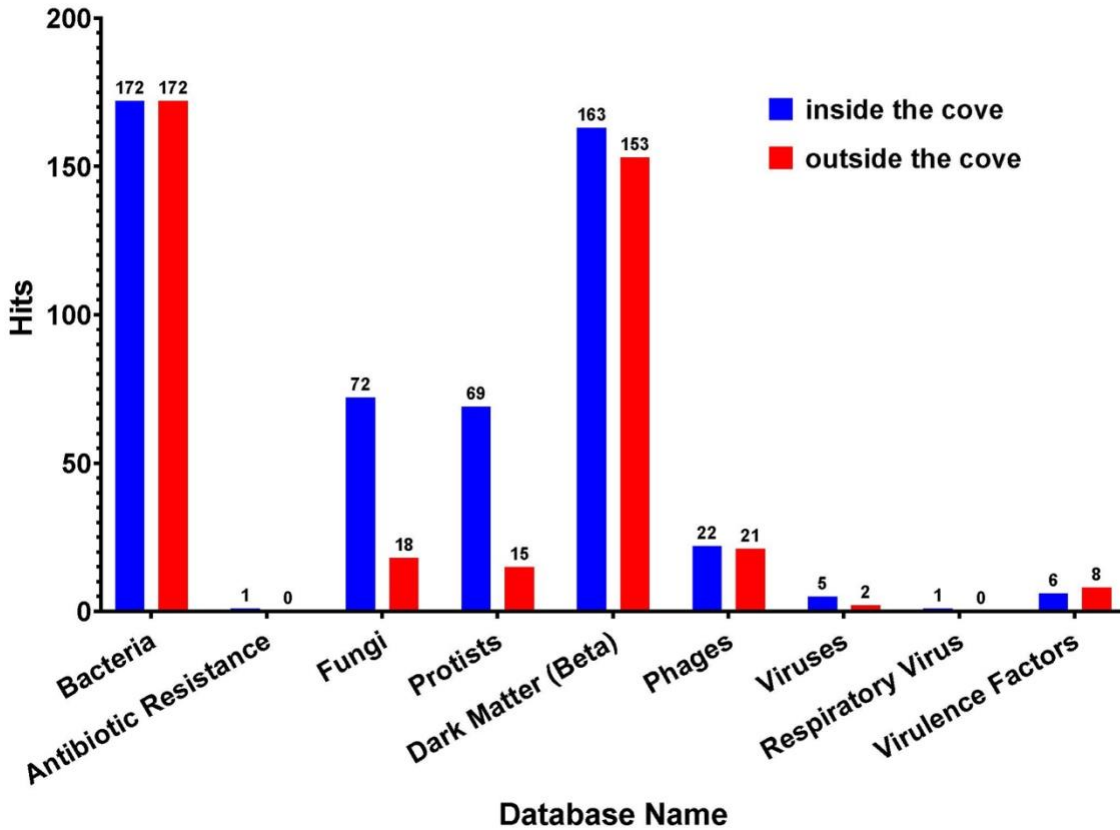


**Figure 17.** YSI-ProDSS field measurements for points inside the cove, from the river outside the cove, and at the entry point of the cove (In front of the cove). Water temperature (°C), conductivity (µS/cm), dissolved oxygen (%), pH, chlorophyll (µg/L), and phycocyanin levels (µg/L, as a proxy for total cyanobacteria).

The two points outside and inside the cove were sampled for full genome sequencing (metagenomics). The DNA libraries were mapped against curated databases that included: bacteria, including those with antibiotic resistance, fungi, protists, phages, viruses, including respiratory viruses, and virulence factors. The dark matter is a database for non-culturable microbes, with poorly curated database that is in its beta stage. The data is represented as relative abundance, and the identified species could be of low or high abundance. The sequencing data identified more species inside the cove than outside the cove (Fig. 18). Full analysis of the sequencing data is included in this report as appendix D.

- Sample 1 in the whole genome sequencing analysis report, Appendix D1, denotes the water sample from inside the cove.
- Sample 2 in the whole genome sequencing analysis report, Appendix D2, denotes the water sample from outside the cove.

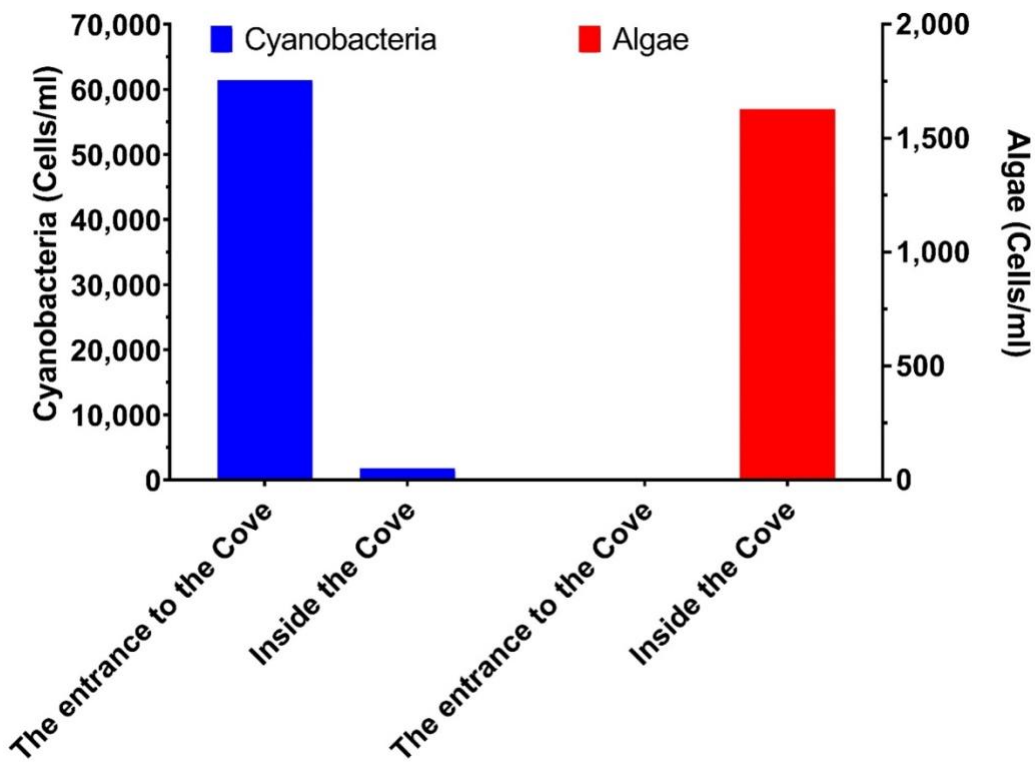
## sequencing analysis



**Figure 18.** Metagenomics analysis for two sample points in site 43, outside and inside the cove, The sequenced DNA libraries were mapped against curated databases that included: bacteria, including those with antibiotic resistance, fungi, protists, phages, viruses, including respiratory viruses, and virulence factors. The dark matter is a database for non-culturable microbes, with poorly curated database that is in its beta stage.

Cell count from samples collected from inside the cove show decreased cyanobacterial cell count compared to a sample collected from the entry point to cove, close to the river, but increased green algae cell count compared to the entry point (Fig. 19)

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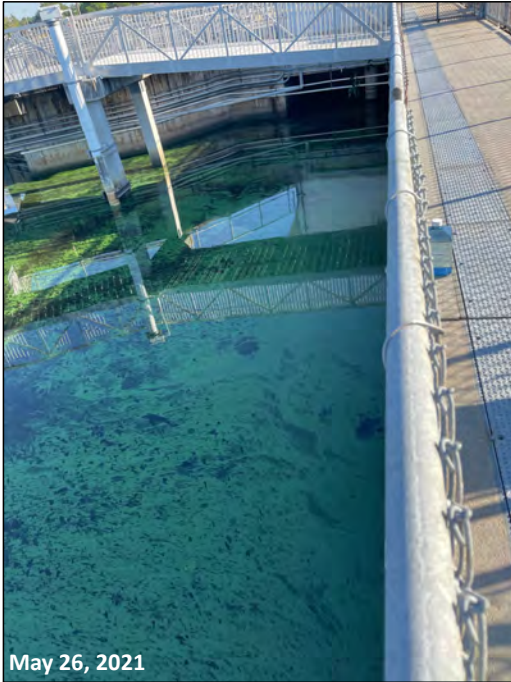
**Figure 19.** Cell count of cyanobacterial cells and green algae cells from a sample collected inside the cove and a sample collected from the entry point, close the river. Cyanobacterial cell count is substantially lower from the sample inside the cove in comparison the entry point, while the green algae cell count is substantially greater from the sample collected inside the cove compared the sample from the entry point.

### 10. Concluding Remarks

The C-43 Canal/ Caloosahatchee River project demonstrated the efficacy and safety of the *Lake Guard*<sup>®</sup> Oxy. The modular treatment protocols allows for the product to treat heavy cyanobacterial blooms as well as maintaining bloom free conditions through preventative treatments. The 15-day project mitigated cyanobacterial blooms and maintained bloom free conditions of residential areas over a 48-mile river segment. Only 5.1 tons out of the 20-ton FL DEP stockpile were used in this project. This relatively small amount of the *Lake Guard*<sup>®</sup> Oxy provided immediate relief to the residents and eliminated the cyanobacterial scum in residential areas along with the associated health risks and unpleasant conditions that accompany cyanobacterial blooms.

*11. Selected before and after pictures*

BEFORE Treatment: May 26, 2021



AFTER Treatment: May 30, 2021



Latitude and longitude: 26.7213, -81.69327  
W.P Franklin Lock & Dam, South Recreation Area

BEFORE Treatment: May 26, 2021

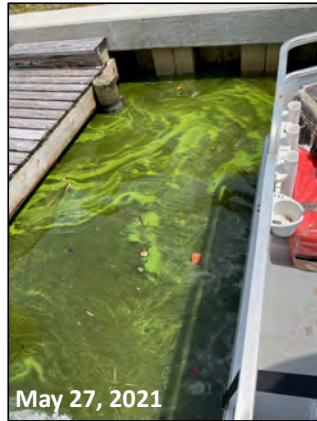
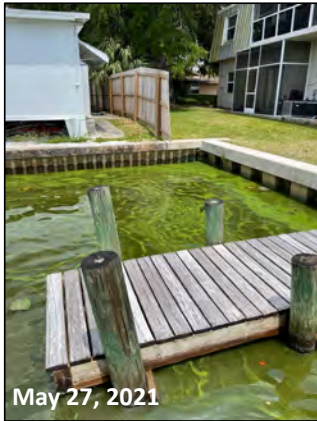


AFTER Treatment: May 28, 2021



Latitude and longitude:  
26.72282, -81.71166

BEFORE Treatment: May 27, 2021



AFTER Treatment: May 28, 2021



Latitude and longitude: 26.72334, -81.71473

Before Treatment: May 28, 2021



May 28, 2021



May 28, 2021

After Treatment: May 31 and June 1, 2021



May 31, 2021



Jun 1, 2021

Latitude and longitude: 26.72487, -81.69367

## Before Treatment: May 28, 2021



## After Treatment: June 1, 2021



Latitude and longitude: 26.72687, -81.69729



BEFORE Treatment: June 4, 2021



AFTER Treatment: June 5, 2021



Latitude and longitude:  
26.74294, -81.51729

## **Appendix A:**

### *An overview of the 56 individual sites treated by BlueGreen/ Modica*

Presented below are the 56 different sites for which 112 treatments and retreatments were applied, using a total amount of 10,260 lb (5.1 tons) of the *Lake Guard*<sup>®</sup> Oxy product. Location, estimated area, aerial photos, site photos, treatment date, time, dosage and total amount, and additional measurement data (if available) are presented for each site.

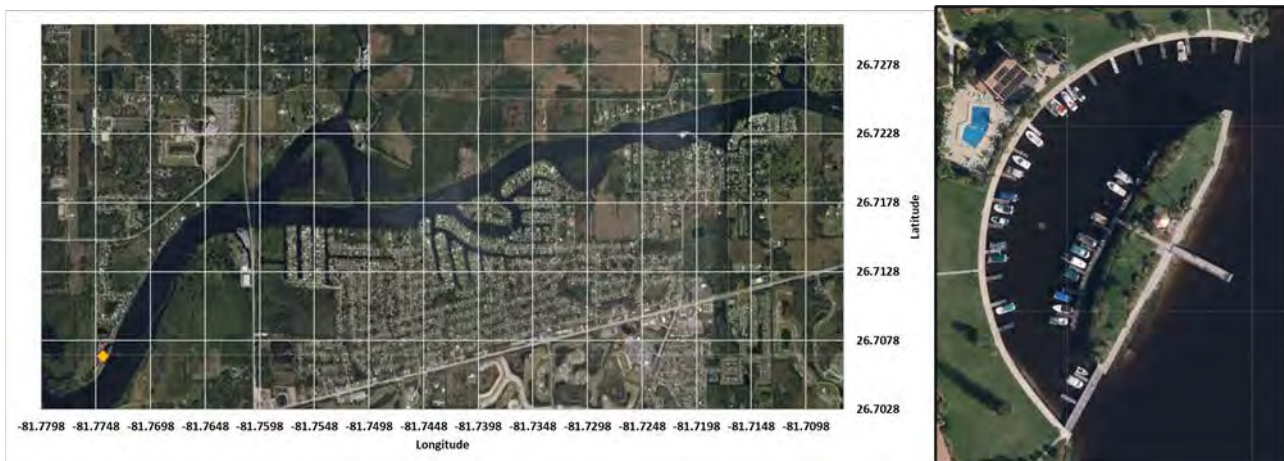
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**Site 1 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
1	26.70664	-81.77408	1.8

**Aerial photographs**



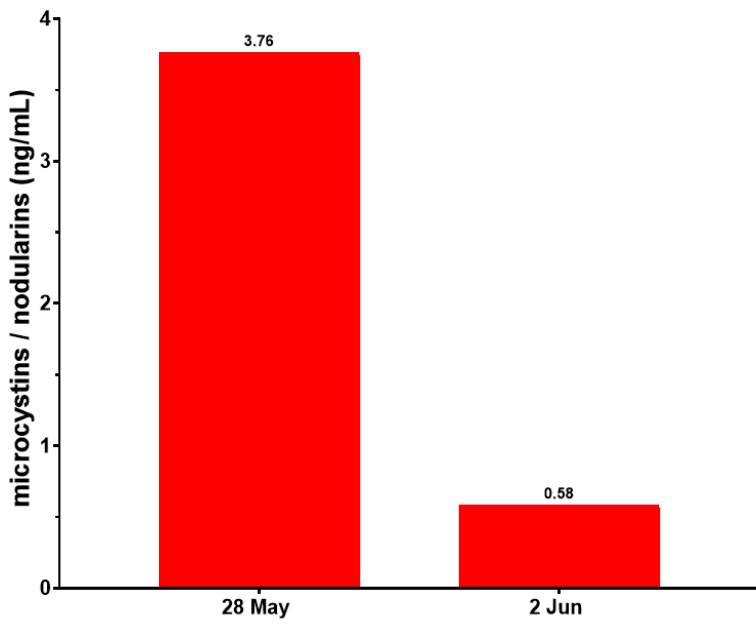
**Treatment details**

date	28 May	1 June	4 June
Treatment (lb)	35	100	25
dose (lb/acre)	19.4	55.6	13.9

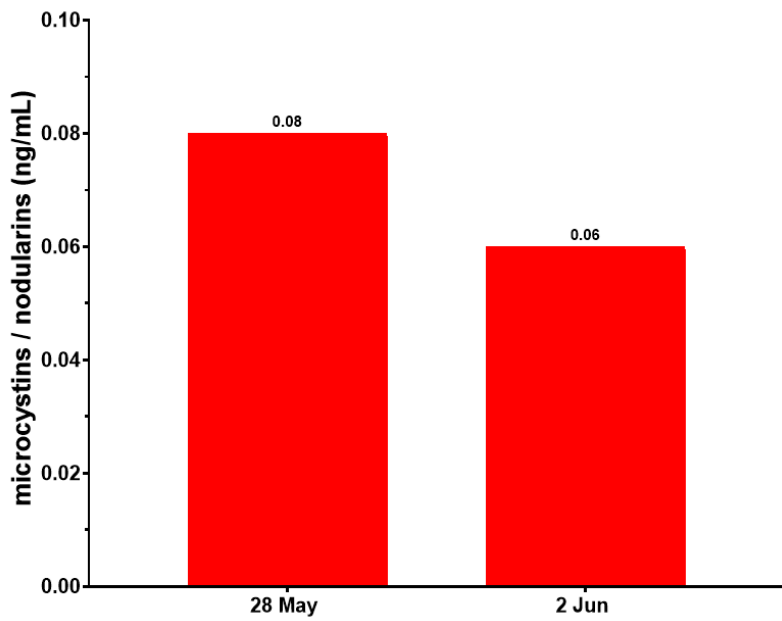
**Site photos**



Site 1 : Microcystins / Nodularins



Site 1 : Saxitoxins



Microcystins / nodularins (top panel) and saxitoxins (bottom panel) (in ng/mL, ppb) found in site #1 on May 28<sup>th</sup> and June 2<sup>nd</sup>, 2021.

## Site 2 – details of the treatment conducted by *BlueGreen/Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
2	26.71293	-81.76067	3.3

### Aerial photographs



### Treatment details

date	28 May
Treatment (lb)	80
dose (lb/acre)	24.2

### Site photos



## Site 3 – details of the treatment conducted by *BlueGreen/Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
3	26.71329	-81.75943	3.2

### Aerial photographs



### Treatment details

date	28 May	7 June
Treatment (lb)	85	5
dose (lb/acre)	26.6	1.6

### Site photos



**Site 4 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
4	26.71275	-81.75729	4.5

**Aerial photographs**



**Treatment details**

date	28 May	29 May
Treatment (lb)	100	150
dose (lb/acre)	22.2	33.3

**Site photos**



**Site 5 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
5	26.71321	-81.75605	5.7

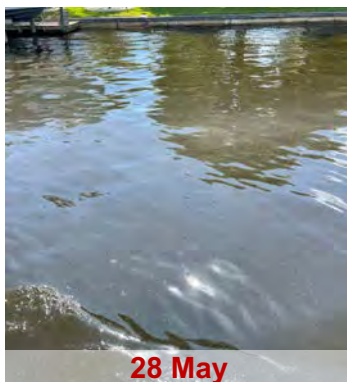
**Aerial photographs**



**Treatment details**

date	28 May	29 May	31 May
Treatment (lb)	135	100	100
dose (lb/acre)	23.7	17.5	17.5

**Site photos**





## Site 6 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
6	26.71319	-81.75472	3.8

### Aerial photographs



### Treatment details

date	28 May	29 May	31 May
Treatment (lb)	110	100	100
dose (lb/acre)	28.9	26.3	26.3

### Site photos



**Site 7 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
7	26.71315	-81.75350	5.2

**Aerial photographs**



**Treatment details**

date	28 May	29 May	30 May	31 May
Treatment (lb)	125	100	200	100
dose (lb/acre)	24.0	19.2	38.5	19.2

**Site photos**

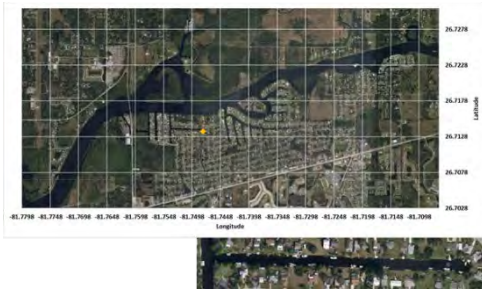


## Site 8 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
8	26.71352	-81.74779	4.9

### Aerial photographs



### Treatment details

date	28 May	30 May
Treatment (lb)	105	150
dose (lb/acre)	21.4	30.6

### Site photos



**Site 9 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
9	26.71985	-81.72520	2.4

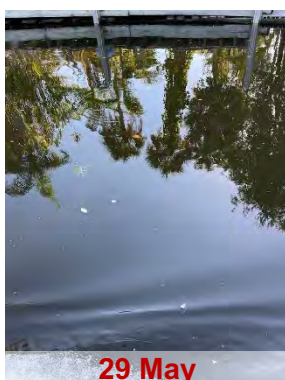
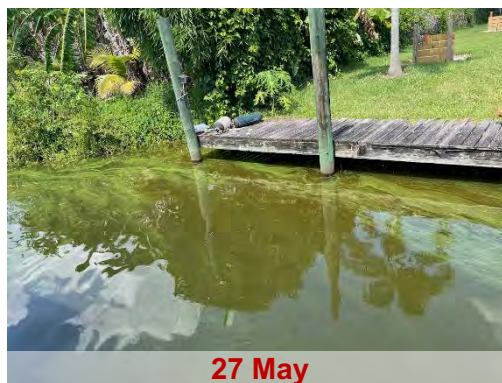
**Aerial photographs**



**Treatment details**

date	28 May	29 May	30 May
Treatment (lb)	95	100	100
dose (lb/acre)	39.6	41.7	41.7

**Site photos**

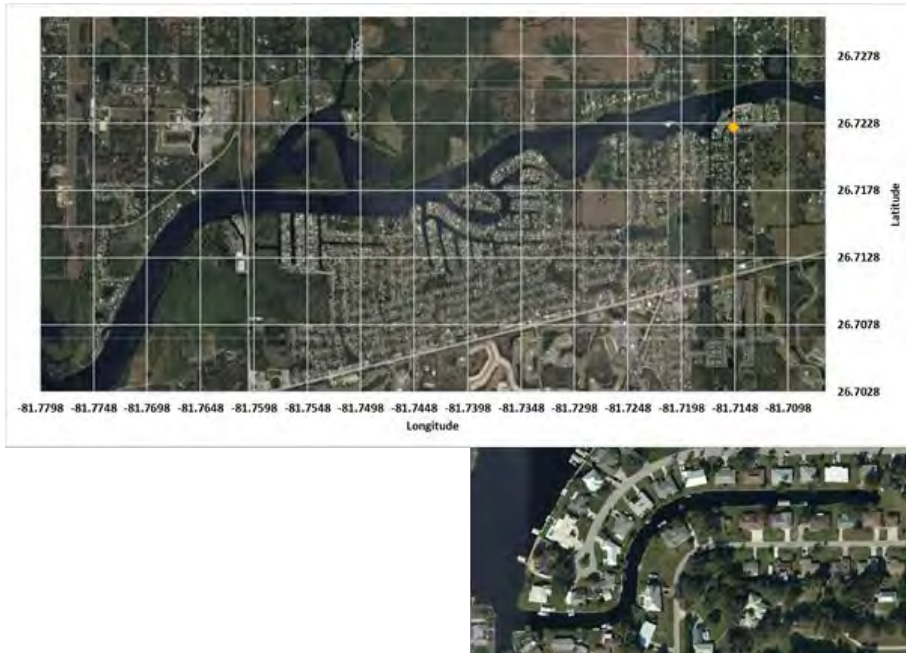


## Site 10 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
10	26.72246	-81.71485	2.2

### Aerial photographs



### Treatment details

date	29 May
Treatment (lb)	100
dose (lb/acre)	45.5

### Site photos



**Site 11 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
11	26.72335	-81.71463	1.1

**Aerial photographs**



**Treatment details**

date	27 May	28 May	29 May	30 May	31 May
Treatment (lb)	65	35	30	200	100
dose (lb/acre)	59.1	31.8	27.3	181.8	90.9

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Site #11 photos



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**Site 12 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
12	26.72288	-81.71145	1.2

**Aerial photographs**



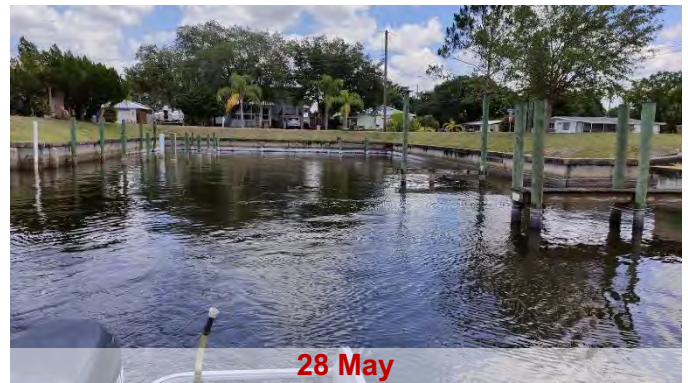
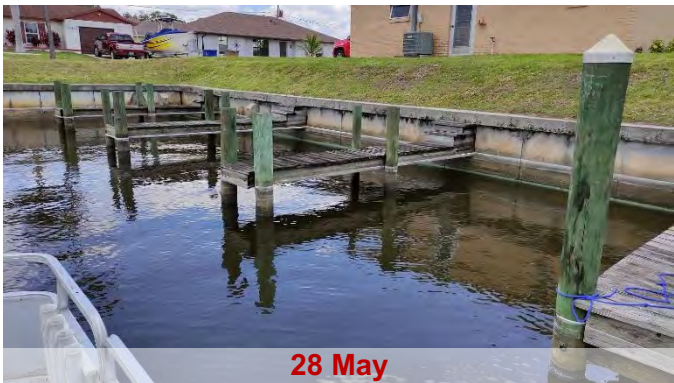
**Treatment details**

date	27 May	28 May	29 May	30 May	31 May
Treatment (lb)	55	30	20	100	100
dose (lb/acre)	45.8	25.0	16.7	83.3	83.3

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**Site #12 photos**



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**Site 13 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
13	26.72825	-81.71017	14

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>29 May</b>
<b>Treatment (lb)</b>	<b>150</b>
<b>dose (lb/acre)</b>	<b>10.7</b>

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**Site 14 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
14	26.72687	-81.69729	2.7

**Aerial photographs**



**Treatment details**

date	28 May	29 May	30 May	1 June	2 June
Treatment (lb)	260	200	100	50	100
dose (lb/acre)	96.3	74.1	37.0	18.5	37.0

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**Site #14 photos**



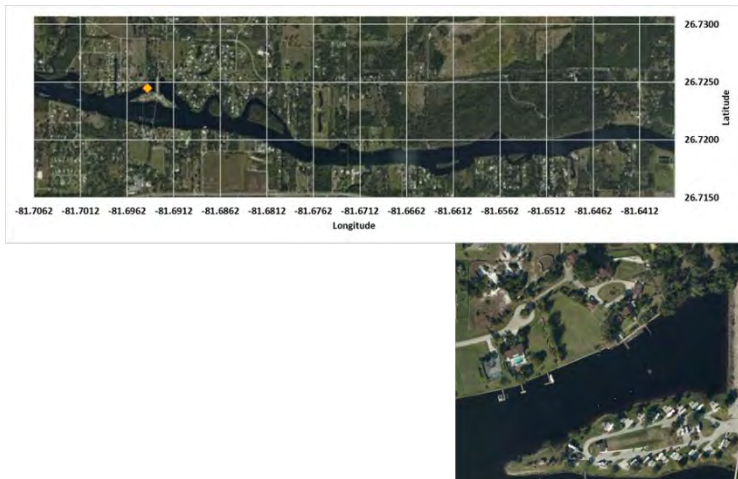
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Site 15 – details of the treatment conducted by *BlueGreen/ Modica*

Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
15	26.72441	-81.69394	9.6

Aerial photographs



Treatment details

date	28 May	29 May	2 June
Treatment (lb)	370	330	150
dose (lb/acre)	38.5	34.4	15.6

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**Site #15 photos**



**28 May**



**28 May**



**29 May**



**1 Jun**



**1 Jun**



**1 Jun**

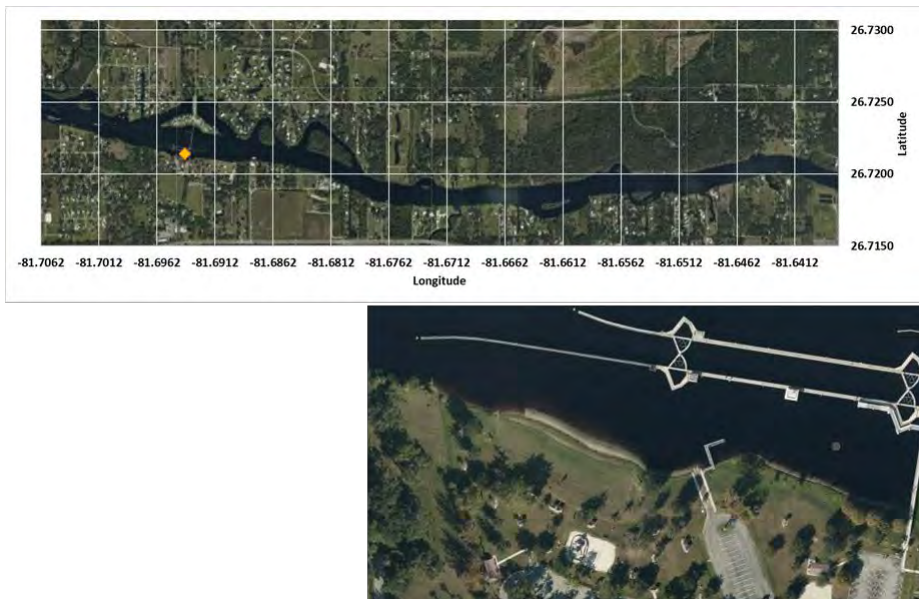
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**Site 16 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
16	26.72135	-81.69373	3.6

**Aerial photographs**

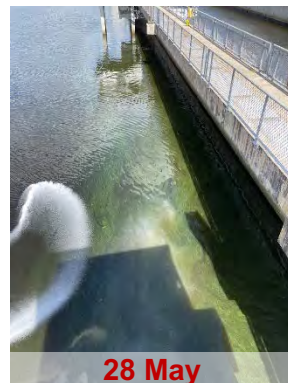


**Treatment details**

date	27 May	28 May	29 May	1 June	2 June	3 June	4 June
Treatment (lb)	130	70	150	25	75	50	5
dose (lb/acre)	36.1	19.4	41.7	6.9	20.8	13.9	1.4

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**Site #16 photos**



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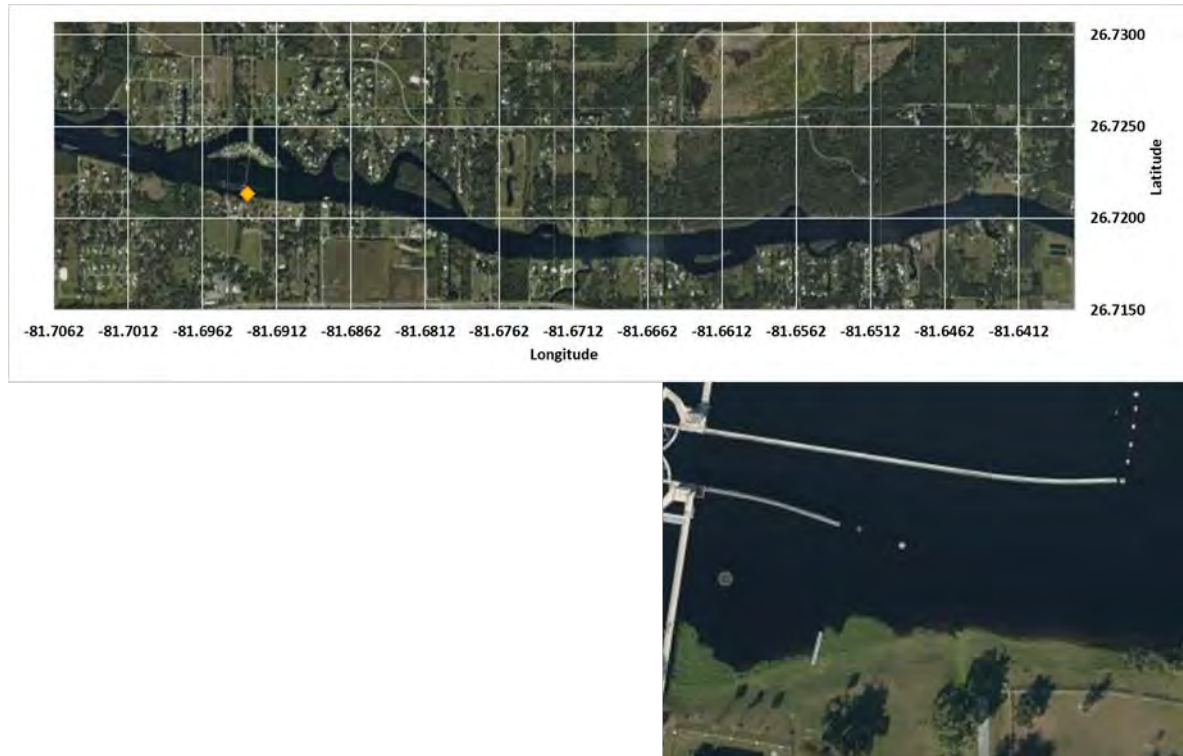


**Site 17 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
17	26.72127	-81.69309	1.2

**Aerial photographs**



**Treatment details**

date	28 May	29 May	1 June	2 June	5 June	10 June
Treatment (lb)	170	100	150	50	27	50
dose (lb/acre)	141.7	83.8	125.0	41.7	22.5	41.7

**Site 18 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
18	26.72437	-81.69175	13.9

**Aerial photographs**



**Treatment details**

date	29 May
Treatment (lb)	150
dose (lb/acre)	10.8

**Site photos**

**Site photos**

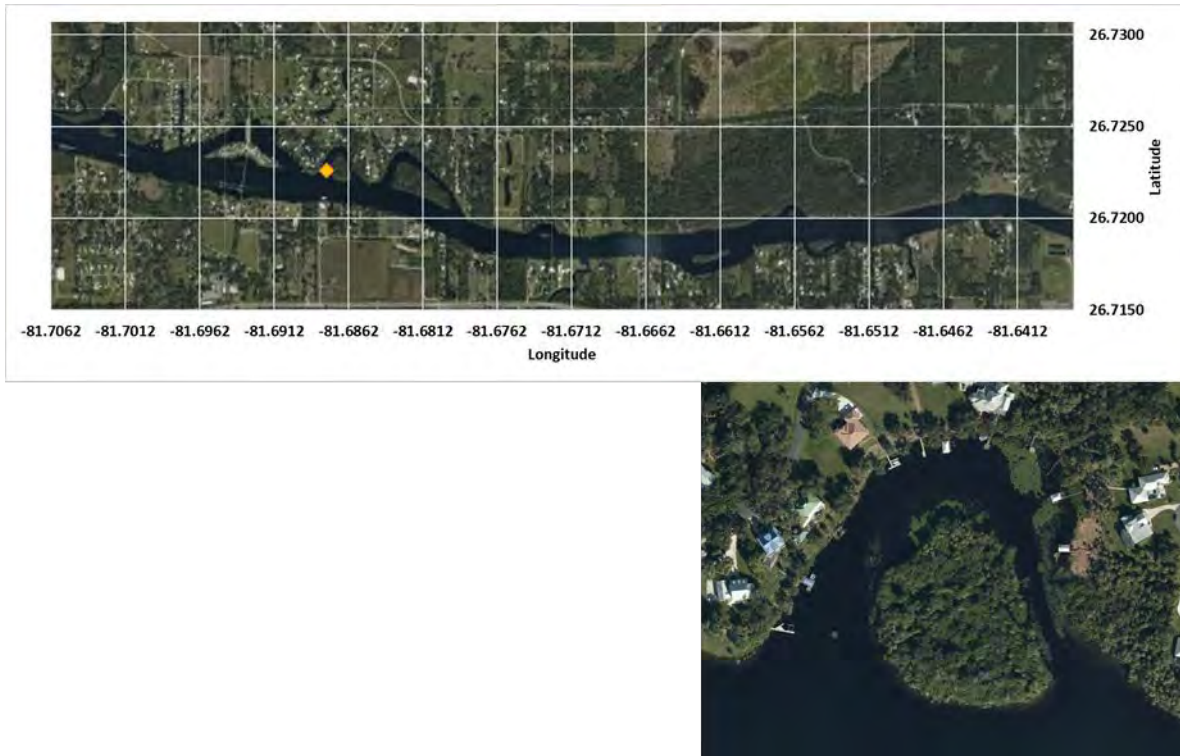


**Site 19 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
19	26.72255	-81.68762	7.6

**Aerial photographs**



**Treatment details**

date	28 May
Treatment (lb)	45
dose (lb/acre)	5.9

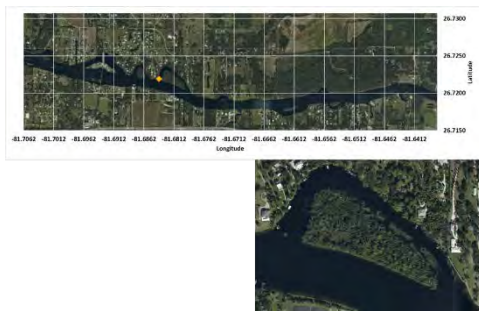
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**Site 20 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
20	26.72187	-81.68363	13.5

**Aerial photographs**



**Treatment details**

date	28 May	8 June
Treatment (lb)	130	30
dose (lb/acre)	9.6	2.2

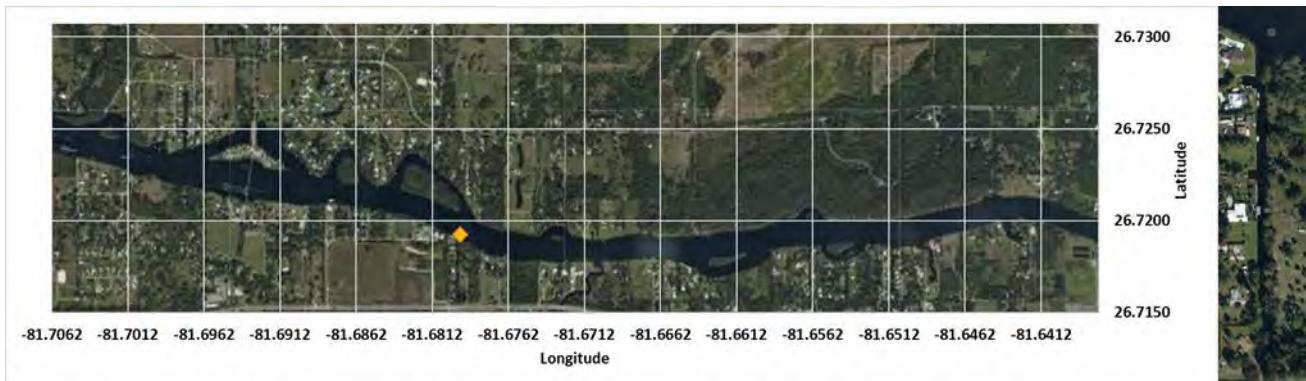
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## Site 21 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
21	26.71920	-81.67930	1.8

### Aerial photographs



### Treatment details

date	28 May
Treatment (lb)	140
dose (lb/acre)	77.8

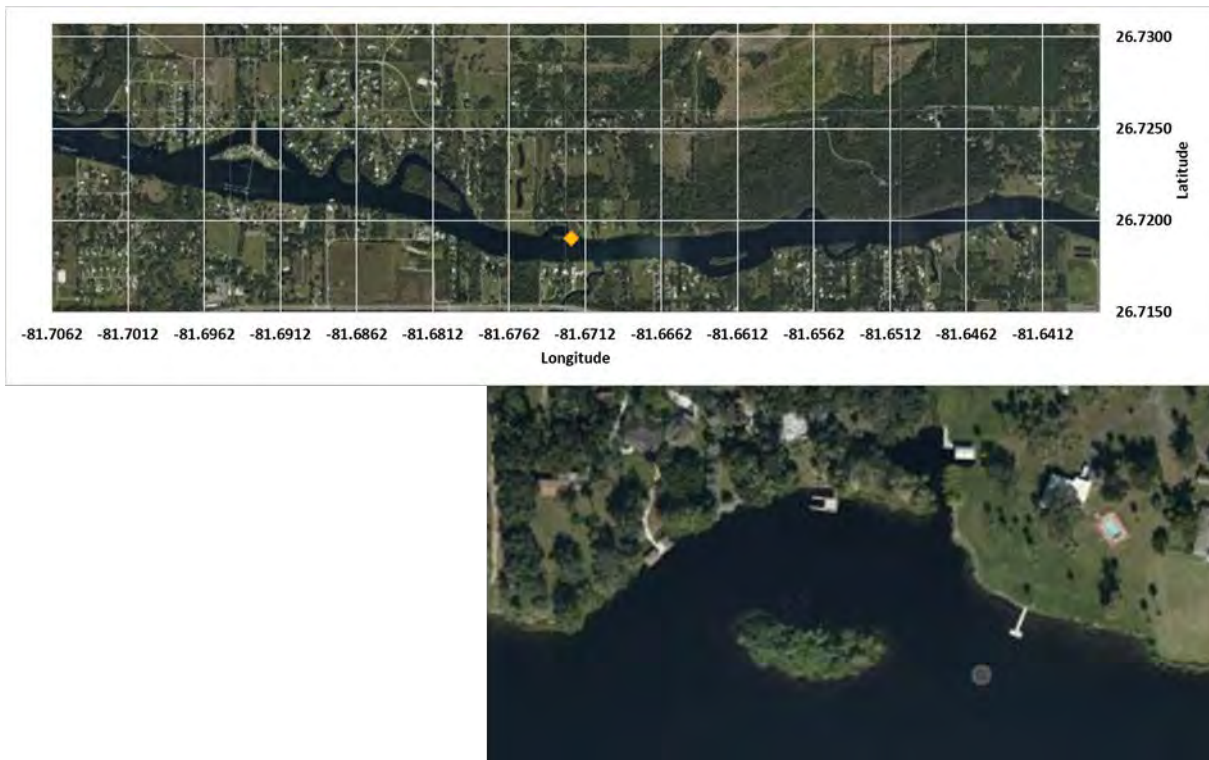
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## Site 22 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
22	26.71898	-81.67206	3.2

### Aerial photographs



### Treatment details

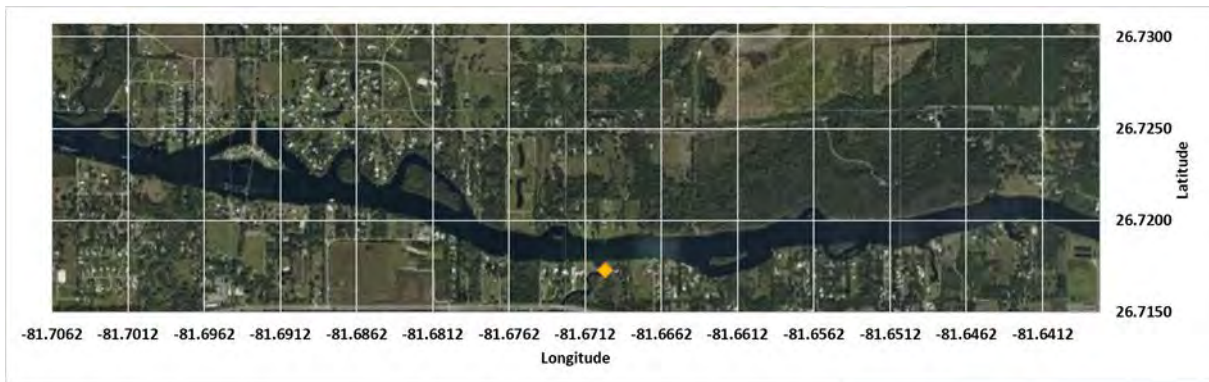
date	28 May
Treatment (lb)	25
dose (lb/acre)	7.8

## Site 23 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
23	26.71721	-81.66987	3.9

### Aerial photographs



### Treatment details

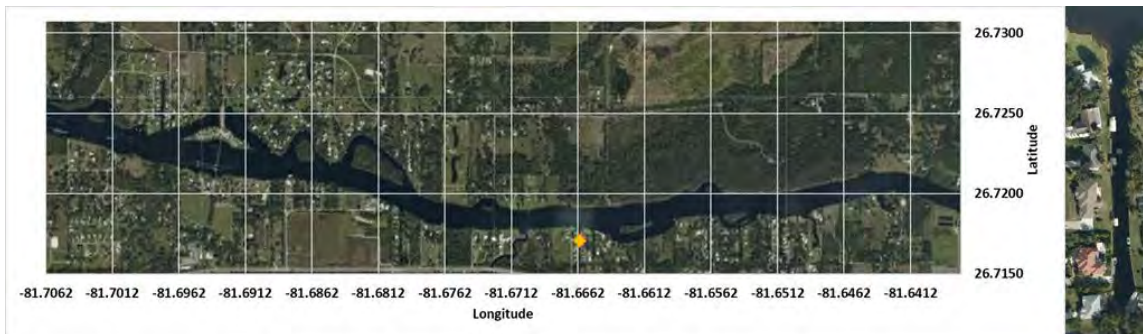
date	28 May
Treatment (lb)	70
dose (lb/acre)	17.9

**Site 24 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
24	26.71699	-81.66603	1

**Aerial photographs**



**Treatment details**

date	28 May
Treatment (lb)	50
dose (lb/acre)	50.0

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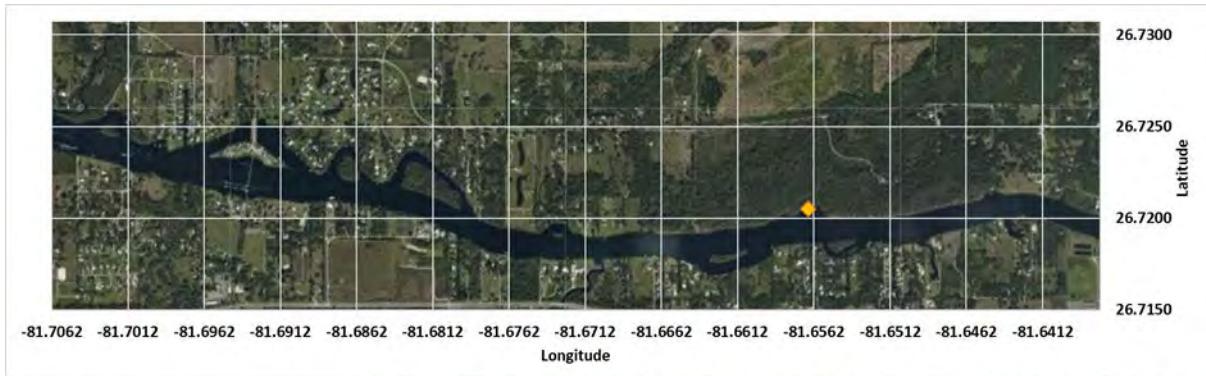


**Site 25 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
25	26.72046	-81.65650	2.6

**Aerial photographs**



**Treatment details**

date	28 May
Treatment (lb)	30
dose (lb/acre)	11.5

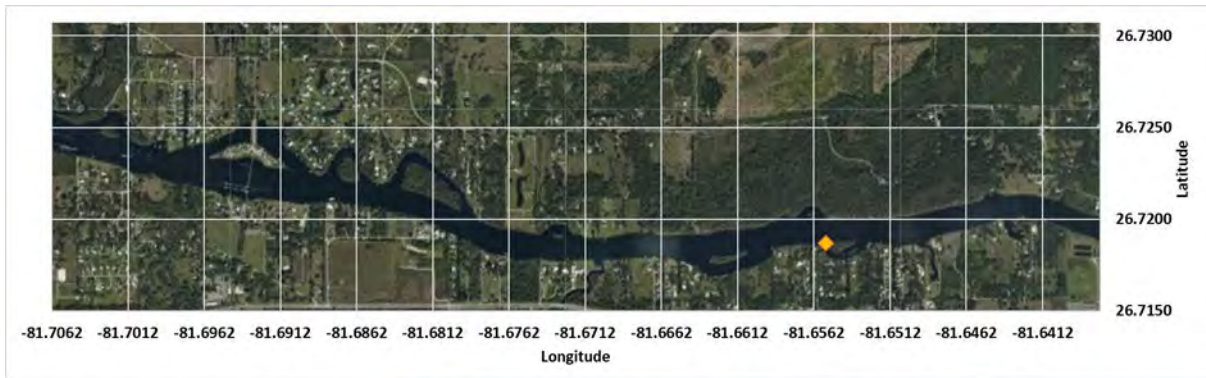
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**Site 26 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
26	26.71863	-81.65537	5.6

**Aerial photographs**



**Treatment details**

date	28 May	8 June
Treatment (lb)	120	10
dose (lb/acre)	21.4	1.8

**Site photos**

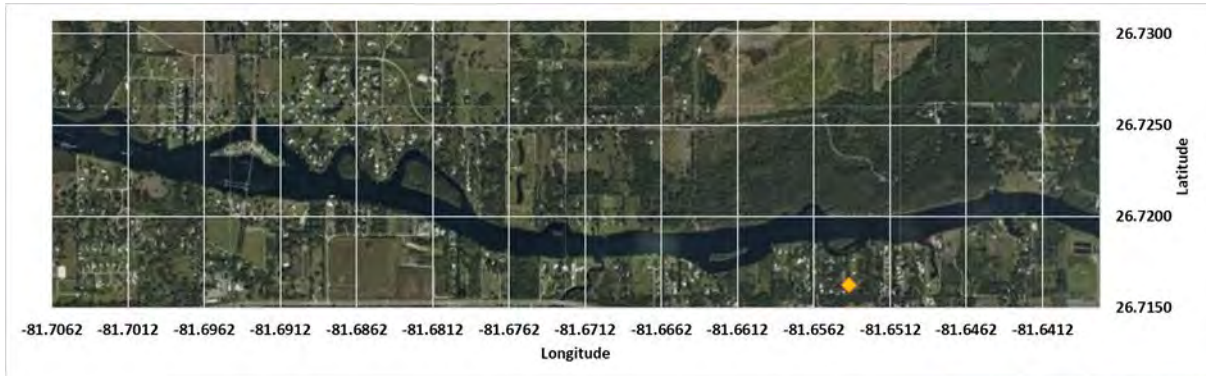


**Site 27 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
27	26.71620	-81.65383	1.3

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>28 May</b>
<b>Treatment (lb)</b>	130
<b>dose (lb/acre)</b>	100.0

**Site photos**

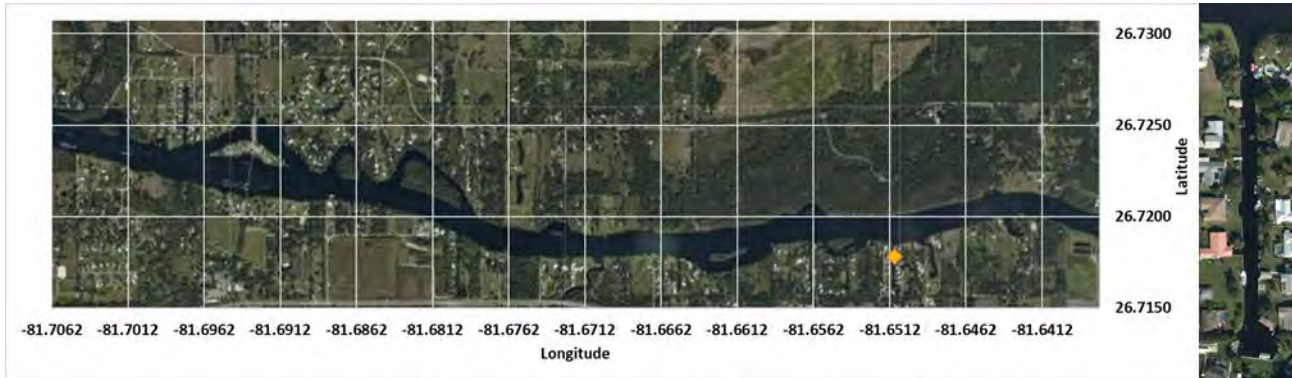


Site 28 – details of the treatment conducted by *BlueGreen/ Modica*

Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
28	26.71775	-81.65085	1.4

Aerial photographs



Treatment details

date	28 May
Treatment (lb)	60
dose (lb/acre)	42.9

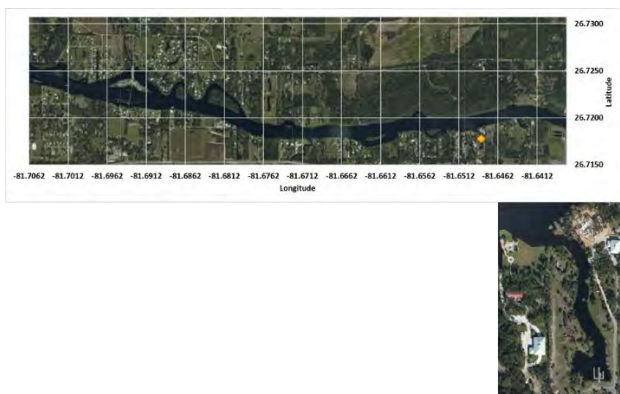
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## Site 29 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
29	26.71772	-81.64829	2

### Aerial photographs



### Treatment details

date	28 May
Treatment (lb)	15
dose (lb/acre)	7.5

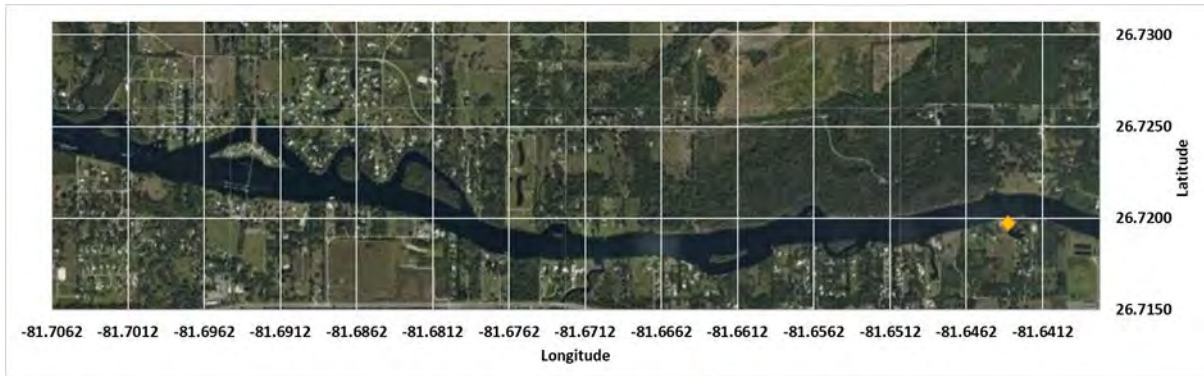
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## Site 30 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
30	26.71968	-81.64342	3.1

### Aerial photographs



### Treatment details

date	28 May
Treatment (lb)	15
dose (lb/acre)	4.8

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**Site 31 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
31	26.71756	-81.63257	2.8

**Aerial photographs**



**Treatment details**

date	8 June
Treatment (lb)	5
dose (lb/acre)	1.8

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**Site 32 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
32	26.71679	-81.62903	3.6

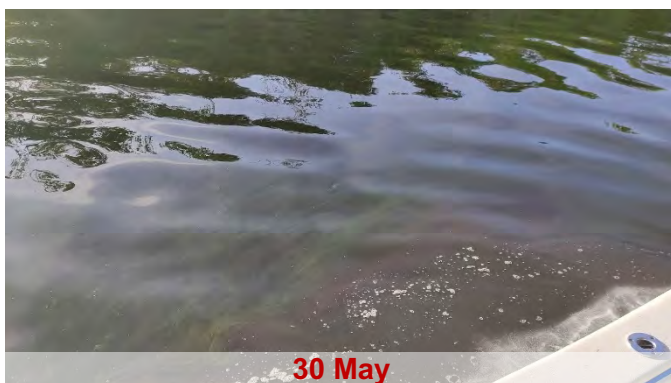
**Aerial photographs**



**Treatment details**

<b>date</b>	<b>30 May</b>
<b>Treatment (lb)</b>	100
<b>dose (lb/acre)</b>	27.8

**Site photos**





**Site 33 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
33	26.71398	-81.62232	1.7

**Aerial photographs**



**Treatment details**

date	29 May
Treatment (lb)	190
dose (lb/acre)	111.8

**Site photos**



**Site 34 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
34	26.71397	-81.62113	1.6

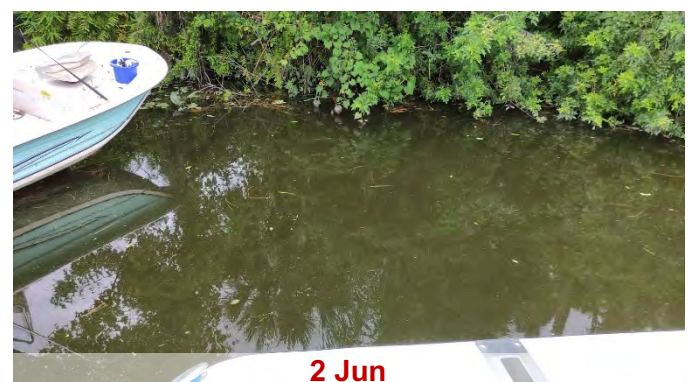
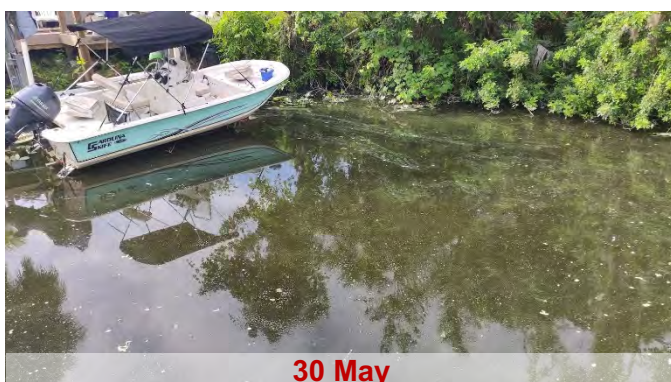
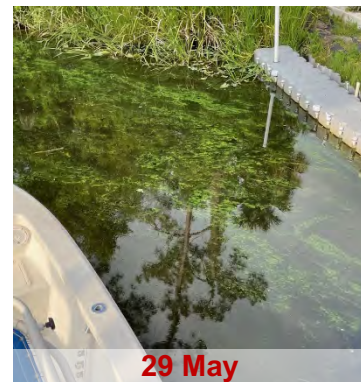
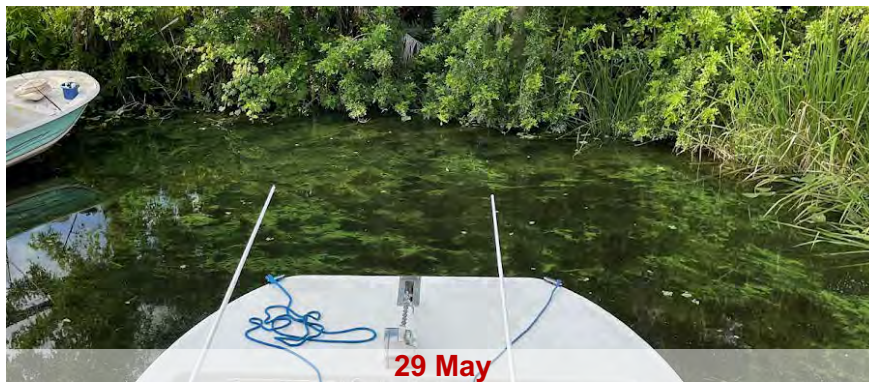
**Aerial photographs**



**Treatment details**

date	29 May
Treatment (lb)	180
dose (lb/acre)	112.5

**Site photos**



**Site 35 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
35	26.71387	-81.61998	1.3

**Aerial photographs**



**Treatment details**

date	29 May	30 May	1 June	7 June	9 June
Treatment (lb)	220	150	200	30	15
dose (lb/acre)	169.2	115.4	153.8	23.1	11.5

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**Site #35 photos**



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**Site 36 – details of the treatment conducted by *BlueGreen***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
36	26.71068	-81.60706	7.5

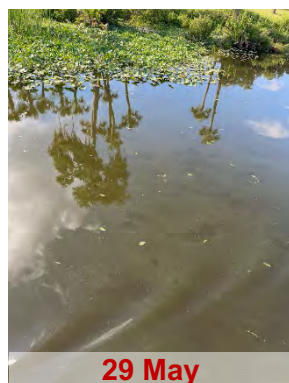
**Aerial photographs**



**Treatment details**

<b>date</b>	<b>29 May</b>
<b>Treatment (lb)</b>	290
<b>dose (lb/acre)</b>	38.7

**Site photos**



**Site 37 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
37	26.71228	-81.60330	2.5

**Aerial photographs**



**Treatment details**

date	29 May
Treatment (lb)	13
dose (lb/acre)	5.2

*Remainder of page intentionally left blank*

**Site 38 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
38	26.71408	-81.59828	0.5

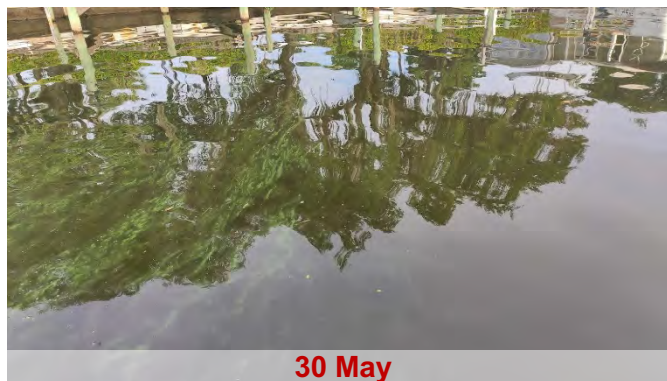
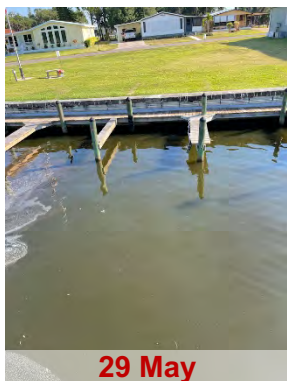
**Aerial photographs**



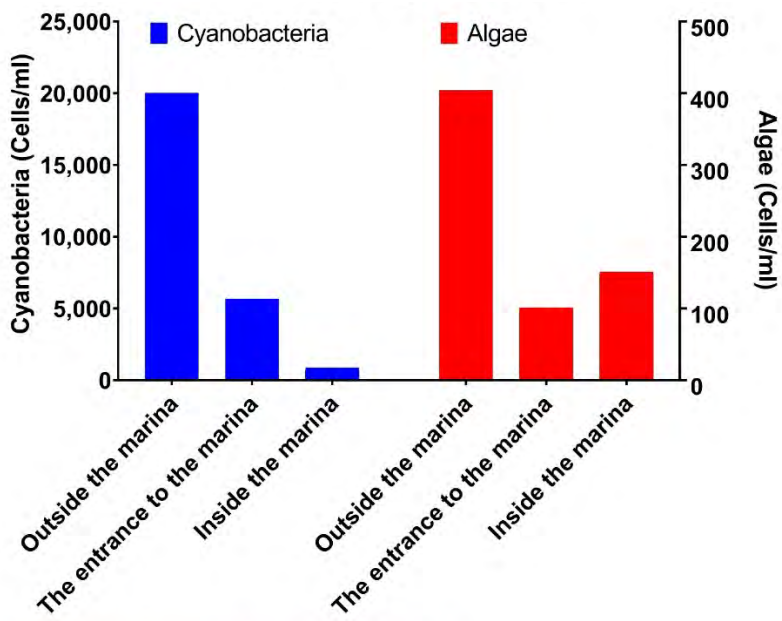
**Treatment details**

date	29 May	9 June
Treatment (lb)	7	10
dose (lb/acre)	14.0	20.0

**Site photos**



**Phytoplankton population composition – Site 38 - for a water sample from May 30<sup>th</sup>, 2021:**



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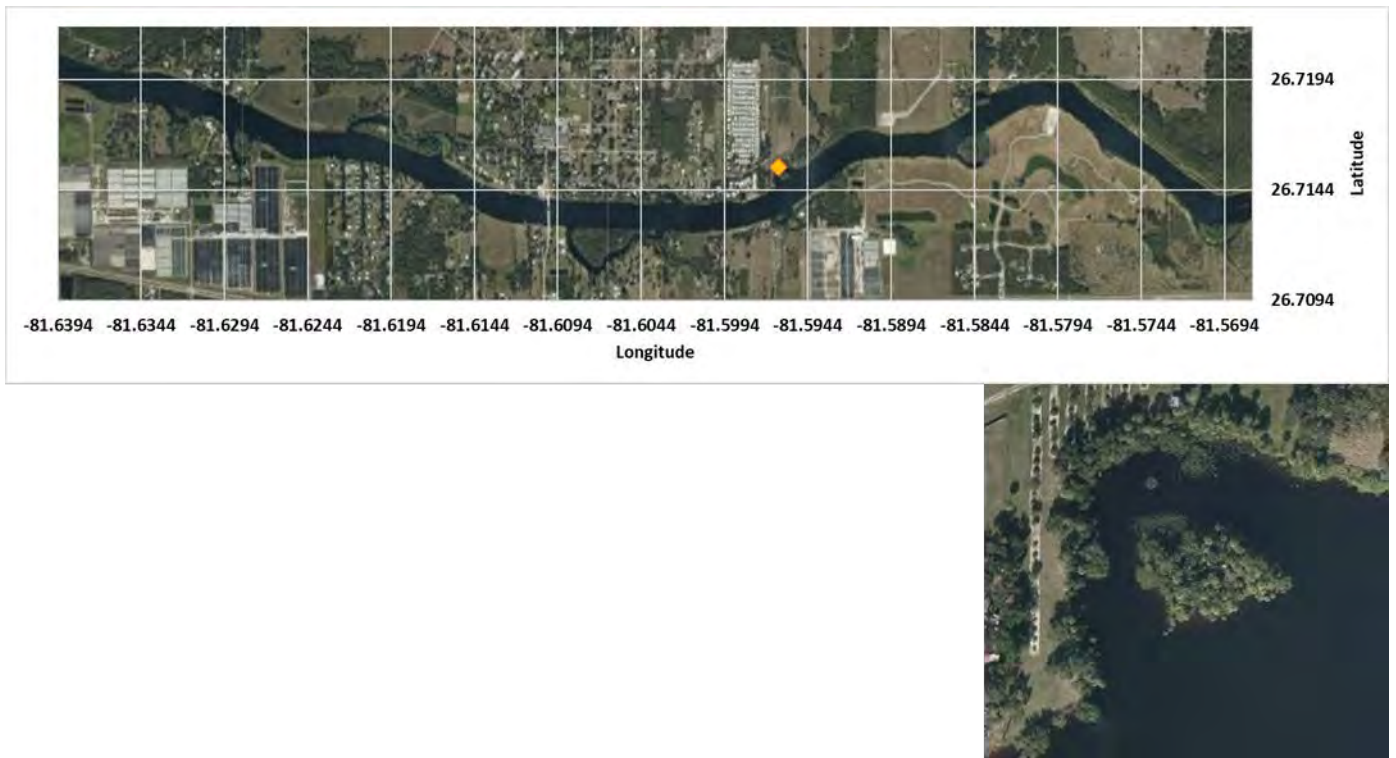


**Site 39 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
39	26.71540	-81.59609	1.9

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>29 May</b>
<b>Treatment (lb)</b>	100
<b>dose (lb/acre)</b>	52.6

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**Site 40 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
40	26.71864	-81.58434	2

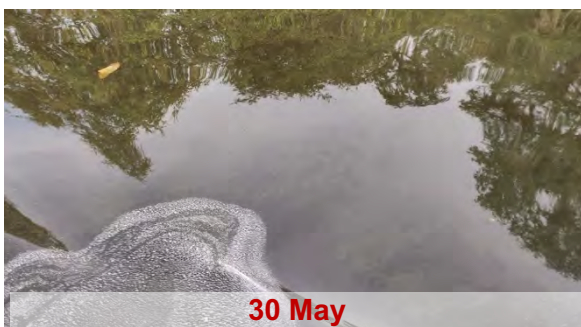
**Aerial photographs**



**Treatment details**

date	30 May
Treatment (lb)	100
dose (lb/acre)	50.0

**Site photos**



30 May

30 May

30 May

Site 41 – details of the treatment conducted by *BlueGreen/ Modica*

Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
41	26.71650	-81.53870	1.2

Aerial photographs



Treatment details

date	4 June	7 June
Treatment (lb)	7	1
dose (lb/acre)	5.8	0.8

## Site 42 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
42	26.74171	-81.51926	2.3

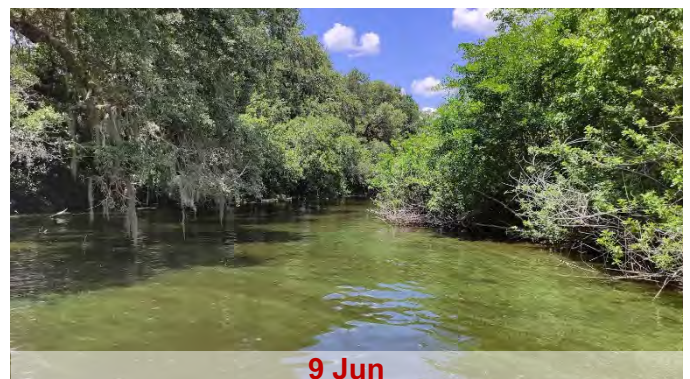
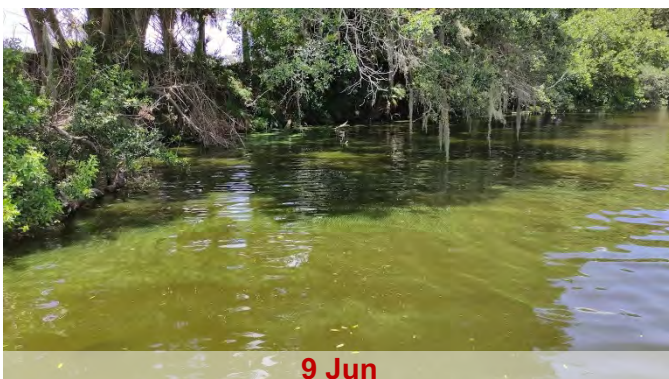
### Aerial photographs



### Treatment details

date	9 June
Treatment (lb)	20
dose (lb/acre)	8.7

### Site photos

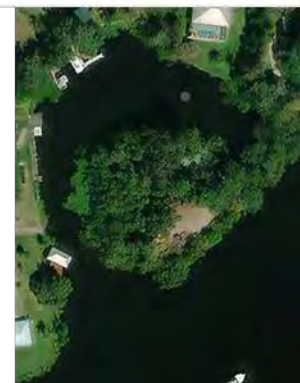


**Site 43 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
43	26.74294	-81.51729	2.3

**Aerial photographs**



**Treatment details**

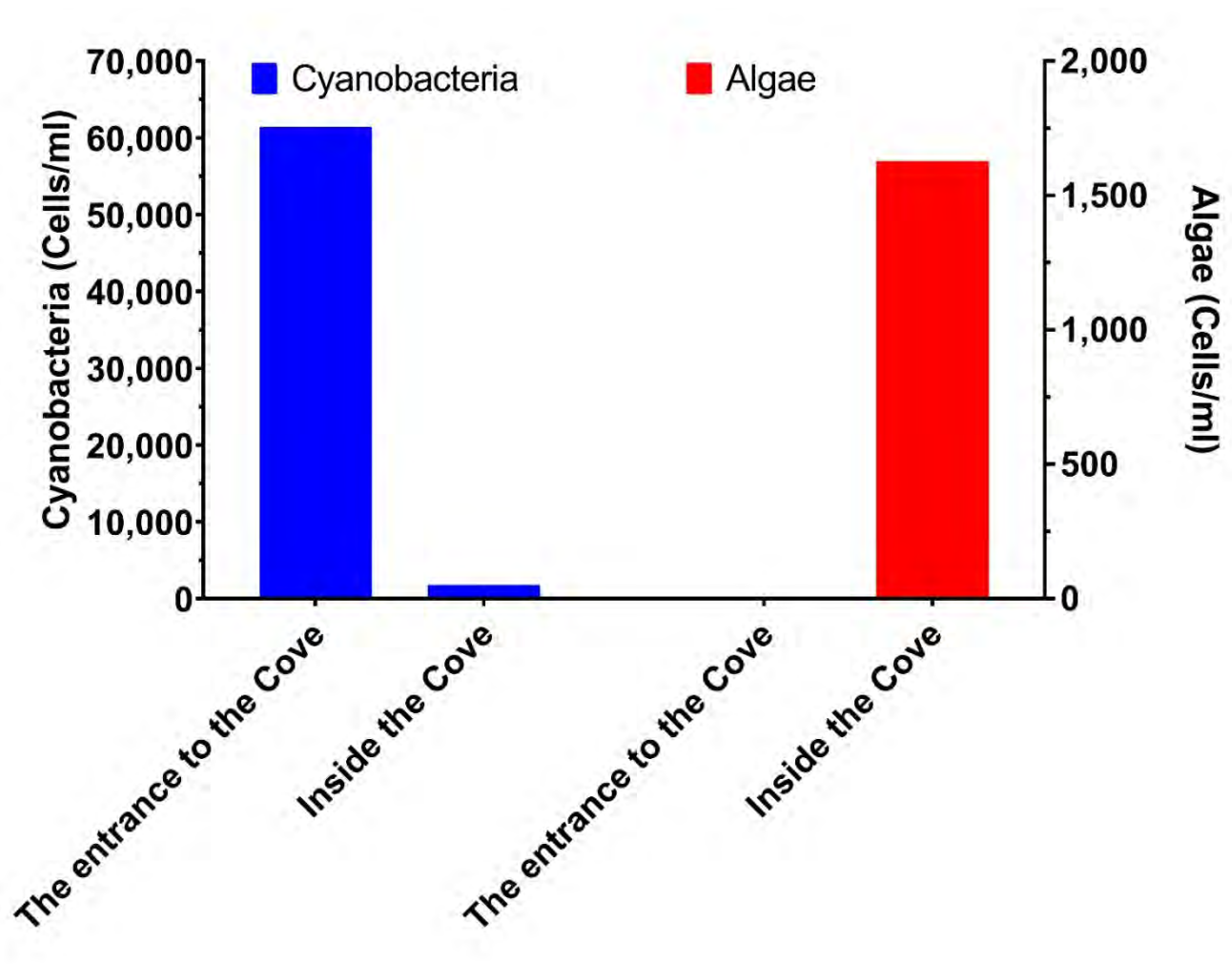
date	4 June	6 June	8 June	10 June
Treatment (lb)	100	230	50	160
dose (lb/acre)	43.5	100	21.7	69.6

Site #43 photos





Phytoplankton population composition – Site 43 - for a water sample obtained on June 6<sup>th</sup>, 2021:



## Site 44 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
44	26.74323	-81.49766	0.6

### Aerial photographs



### Treatment details

date	5 June
Treatment (lb)	25
dose (lb/acre)	41.7

### Site photos





**Site 45 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
45	26.75222	-81.46245	1.5

**Aerial photographs**



**Treatment details**

date	7 June
Treatment (lb)	50
dose (lb/acre)	33.3

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**Site 46 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
46	26.77136	-81.43512	3.9

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>7 June</b>
<b>Treatment (lb)</b>	40
<b>dose (lb/acre)</b>	10.3

**Site photos**



**Site 47 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
47	26.76756	-81.42937	0.7

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>7 June</b>
<b>Treatment (lb)</b>	<b>75</b>
<b>dose (lb/acre)</b>	<b>107.1</b>

**Site 48 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
48	26.76653	-81.42513	0.6

**Aerial photographs**



**Treatment details**

<b>date</b>	<b>8 June</b>
<b>Treatment (lb)</b>	<b>50</b>
<b>dose (lb/acre)</b>	<b>83.3</b>

Site 49 – details of the treatment conducted by *BlueGreen/ Modica*

Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
49	26.77087	-81.39178	11.5

Aerial photographs



Treatment details

date	8 June
Treatment (lb)	15
dose (lb/acre)	1.3



**Site 50 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
50	26.79575	-81.29435	3.2

**Aerial photographs**

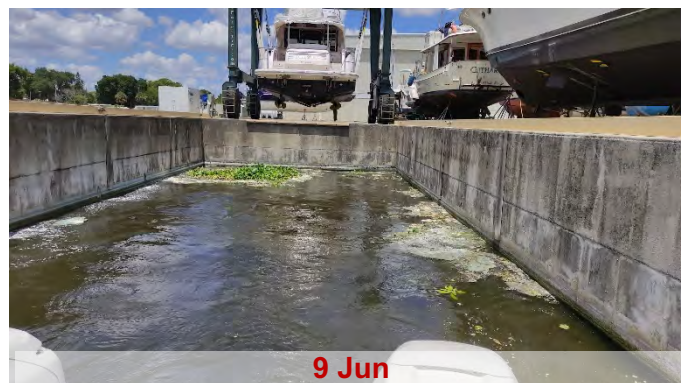


**Treatment details**

date	7 June	9 June
Treatment (lb)	155	120
dose (lb/acre)	48.4	37.5

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Site #50 photos





**Site 51 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
51	26.79428	-81.29254	1.3

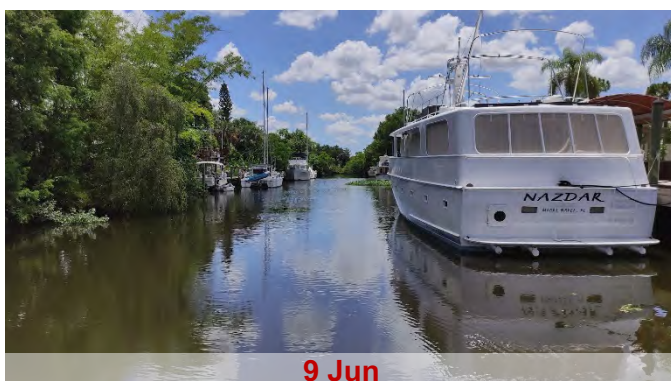
**Aerial photographs**



**Treatment details**

date	7 June
Treatment (lb)	40
dose (lb/acre)	30.8

**Site photos**



## Site 52 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
52	26.797364	-81.291727	1

### Aerial photographs



### Treatment details

date	9 June
Treatment (lb)	5
dose (lb/acre)	5.0

### Site photos



**Site 53 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
53	26.79436	-81.29136	1.6

**Aerial photographs**



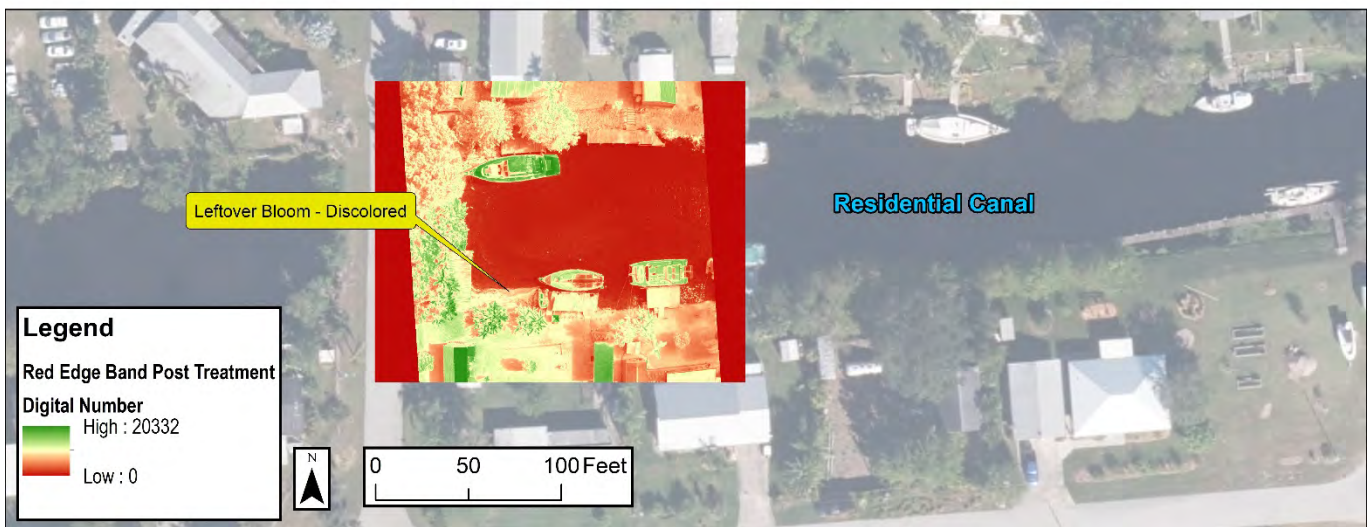
**Treatment details**

date	7 June	9 June
Treatment (lb)	160	120
dose (lb/acre)	100.0	75.0

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Site #53 photos





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## Site 54 – details of the treatment conducted by *BlueGreen/ Modica*

### Location and estimated area

# Site	Latitude	Longitude	Estimated area (acre)
54	26.79643	-81.29040	1.3

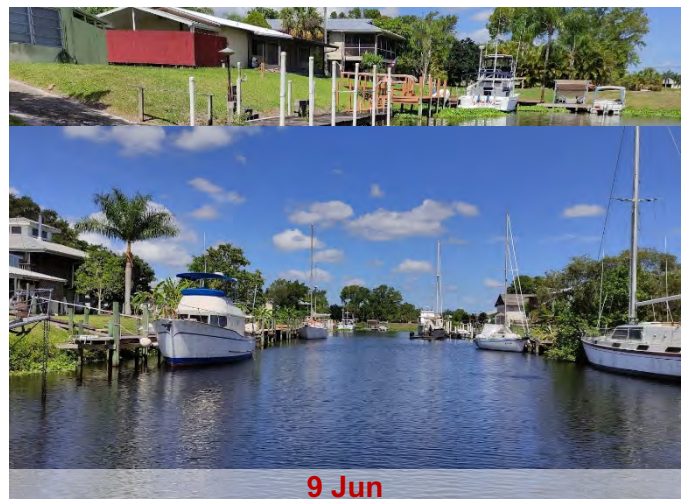
### Aerial photographs



### Treatment details

date	7 June	9 June
Treatment (lb)	40	10
dose (lb/acre)	30.8	7.7

### Site photos



**Site 55 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
55	26.79546	-81.29004	2

**Aerial photographs**

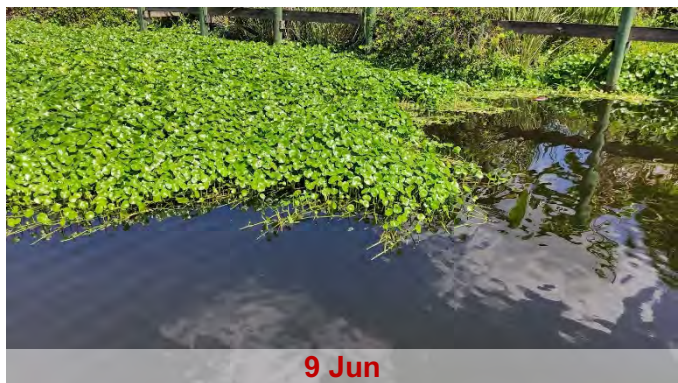
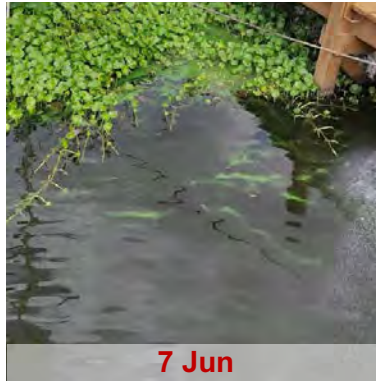


**Treatment details**

date	7 June	9 June
Treatment (lb)	60	20
dose (lb/acre)	30.0	10.0

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Site #55 photos





**Site 56 – details of the treatment conducted by *BlueGreen/ Modica***

**Location and estimated area**

# Site	Latitude	Longitude	Estimated area (acre)
56	26.79382	-81.28939	4.7

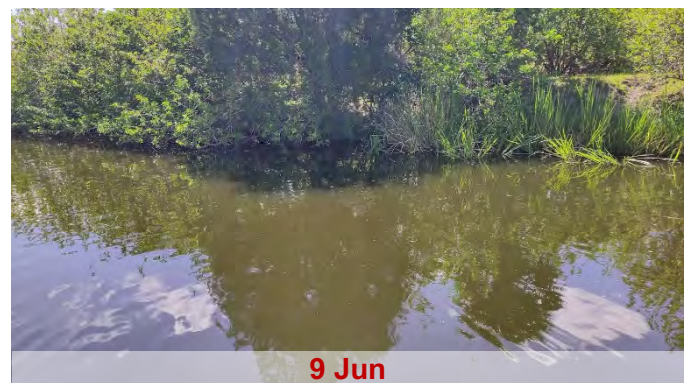
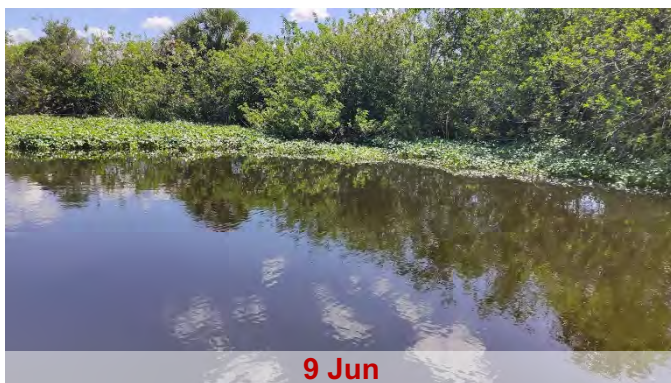
**Aerial photographs**



**Treatment details**

date	7 June	9 June
Treatment (lb)	60	20
dose (lb/acre)	12.8	4.3

**Site photos**



**Anatoxin-a, BMAA, Cylindrospermopsin,  
Microcystins/Nodularins, & Saxitoxins Report**  
*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 5.1 °C upon arrival  
 Report# 210528\_BlueGreen  
 Date Prepared: 4 June 2021  
 Prepared by: Kamil Cieslik

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
1	W.P. Franklin	28 May 2021
2	W.P. Franklin	28 May 2021
3	W.P. Franklin	28 May 2021
4	W.P. Franklin	28 May 2021
5	W.P. Franklin	28 May 2021
9	Caloosahatchee River	28 May 2021

**Toxins** – Anatoxin-a (ATX),  $\beta$ -N-methylamino-L-alanine (BMAA), Cylindrospermopsin (CYN), Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

**Abbreviations**

NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Regent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *ATX/CYN/BMAA/STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *ATX & CYN*

High performance chromatography coupled with tandem mass spectrometry was used for a targeted anatoxin-a and cylindrospermopsin analysis. The  $[M+H]^+$  ion for ATX ( $m/z$  166) was fragmented and the product ions ( $m/z$  91, 131, 149) were monitored. The  $[M+H]^+$  ion for CYN ( $m/z$  416) was fragmented and the product ions ( $m/z$  194, 274, 336) were monitored. The  $[M+H]^+$  ion for the internal standard [ $^{13}C_4$ ]ATX ( $m/z$  171) was fragmented and the product ion ( $m/z$  153) was monitored. The  $[M+H]^+$  ion for the internal standard [ $^{15}N_5$ ]CYN (421  $m/z$ ) was fragmented and the product ion ( $m/z$  341) was monitored. The internal standard method was utilized for quantification.

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

### Quality Control

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	2	LFSM	111%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	71 ± 13%
BMAA	50	1	LFSM	110%
BAMA	50	1	LFSM	101%
2,4-DAB	50	1	LFSM	8% <sup>N</sup>
AEG	50	1	LFSM	88%
3,4-DAB	50	1	LFSM	79%
BMAA	50	9	LFSM	114%
BAMA	50	9	LFSM	98%
2,4-DAB	50	9	LFSM	10% <sup>N</sup>
AEG	50	9	LFSM	119%
3,4-DAB	50	9	LFSM	58% <sup>N</sup>
<i>d</i> <sub>3</sub> -BMAA	50	all aliquots	IS	91 ± 3%
CYN	0.1	2	LFSM	102%
[ <sup>15</sup> N <sub>5</sub> ]CYN	1.0	all aliquots	IS	79 ± 2%
ATX	0.1	2	LFSM	128%
[ <sup>13</sup> C <sub>4</sub> ]ATX	1.0	all aliquots	IS	60 ± 34%
STX	0.2	1	LFSM	103%
STX	0.2	2	LFSM	93%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ±50%; IS ± 50%

*N Qualifier*: The low return for 2,4-DAB is likely due to the low response at the analyte concentration that was spiked. All MDLs/MRLs reflect spike returns.

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received

Table 3: The percent reproducibility (%RPD) between lab duplicates or LFSM/LFSMDs prepared and analyzed.

QC Type	Sample ID	Analyte	Value 1	Value 2	% RPD	Pass/Fail (<40%)
LDs	5	MMPB	0.86	0.58	39.6%	Pass

*%RPD is NA when values are below the method reporting/detection limit*

Table 4: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
1	STX	1	0.08	2.7%	0.08	<b>0.08</b>
		1	0.08			
2	STX	1	0.11	2.2%	0.11	<b>0.11</b>
		1	0.11			
3	STX	1	0.08	9.0%	0.08	<b>0.09</b>
		1	0.09			
4	STX	1	0.08	1.8%	0.08	<b>0.09</b>
		1	0.09			
5	STX	1	0.09	2.9%	0.09	<b>0.09</b>
		1	0.09			
9	STX	1	0.08	3.3%	0.08	<b>0.08</b>
		1	0.08			

Table 5: STX-ELISA Quality Control Value Table

Date Analyzed:	4 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.997	≥0.98	PASS
<b>%CV range STDs:</b>	0.1-2.1%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	98%	±40% True Value	PASS
<b>%CV range LFB:</b>	3.0%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	90%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS

Table 6: STX-ELISA Quality Control Value Table

Date Analyzed:	11 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.999	≥0.98	PASS
<b>%CV range STDs:</b>	0.3-5.0%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	78%	±40% True Value	PASS
<b>%CV range LFB:</b>	3.7%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	80%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS

## Summary of Results

Table 7: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method, anatoxin-a (ATX), cylindrospermopsin (CYN),  $\beta$ -N-methylamino-L-alanine (BMAA), and saxitoxins (STX). All data is reported as ng/mL (ppb).

Sample	MCs/NODs (ng/mL)	ATX (ng/mL)	CYN (ng/mL)	BMAA (ng/mL)	STX (ng/mL)
1	8.20	—	—	ND	0.08
2	2.33	ND	ND	ND	0.11
3	2.44	—	—	ND	0.09
4	0.91	—	—	ND	0.09
5	0.72 ± 0.20	—	—	ND	0.09
9	3.76	ND	ND	ND	0.08
<i>MRL (ng/mL):</i>	0.06	0.05	0.05	5.0	0.05
<i>Analyst Initials:</i>	AF	MA	MA	AF	KC
<i>Date Analyzed:</i>	6/14/2021	6/4/2021	6/4/2021	6/7/2021	6/4/2021 6/11/2021

### Interpretations:

Anatoxin-a, cylindrospermopsin, and BMAA were below method detection limits in all samples tested. Microcystins/nodularins were above the current 'EPA Recommended Value for Recreational Criteria and Swimming Advisory' of 8.0 ng/mL (ppb) (EPA, 2019) in the 1 – WP Franklin sample. Saxitoxins were detected in all samples at sub-ppb levels using a saxitoxin specific ELISA.

Submitted by:

  
Mark T. Aubel, Ph.D.  
Lab Director

Date:

June 18, 2021

*The results in this report relate only to the samples listed above.  
This report shall not be reproduced except in full without written approval of the laboratory.*

## Potentially Toxicogenic (PTOX) Cyanobacteria Report

Project: BlueGreen Water Tech

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 5.1 °C upon arrival  
 Report# 210528\_PTOX\_BlueGreen  
 Date Prepared: 2 June 2021  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
1	W.P. Franklin	5/28/21
2	W.P. Franklin	5/28/21
3	W.P. Franklin	5/28/21
4	W.P. Franklin	5/28/21
5	W.P. Franklin	5/28/21
9	Caloosahatchee River	5/28/21

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 1

The potentially toxicogenic (PTOX) cyanobacteria *Cuspidothrix* sp. ( $\geq 15$  filaments per mL), *Dolichospermum* sp. ( $\geq 10$  partial filaments per mL), *Microcystis* sp. ( $\geq 9$  colonies per mL), *Raphidiopsis* sp. ( $> 5$  filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $> 5$  filaments per mL), and *Pseudanabaena* sp. ( $< 100$  filaments per mL) were observed.

## 2

The PTOX cyanobacteria *Cuspidothrix* sp. (>20 filaments per mL), *Dolichospermum* sp. (>15 partial filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>15 filaments per mL), *Microcystis* sp. (≥10 colonies per mL), *Raphidiopsis* sp. (>5 filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

## 3

The PTOX cyanobacteria *Dolichospermum* sp. (>20 filaments per mL), *Cuspidothrix* sp. (>15 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (>10 colonies per mL), *Raphidiopsis* sp. (>5 filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

## 4

The PTOX cyanobacteria *Cuspidothrix* sp. (≥30 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (≥7 colonies per mL), *Raphidiopsis* sp. (>5 filaments per mL), *Dolichospermum* sp. (≥3 partial filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

## 5

The PTOX cyanobacteria *Cuspidothrix* sp. (>30 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (≥7 colonies per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Raphidiopsis* sp. (≥5 filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

## 9

The PTOX cyanobacteria *Dolichospermum* sp. (>15 partial filaments per mL), *Microcystis* sp. (≥7 colonies per mL), *Cuspidothrix* sp. (>5 filaments per mL), cf. *Aphanizomenon* sp. (≥5 filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

### Potential toxin producing genera observed include:

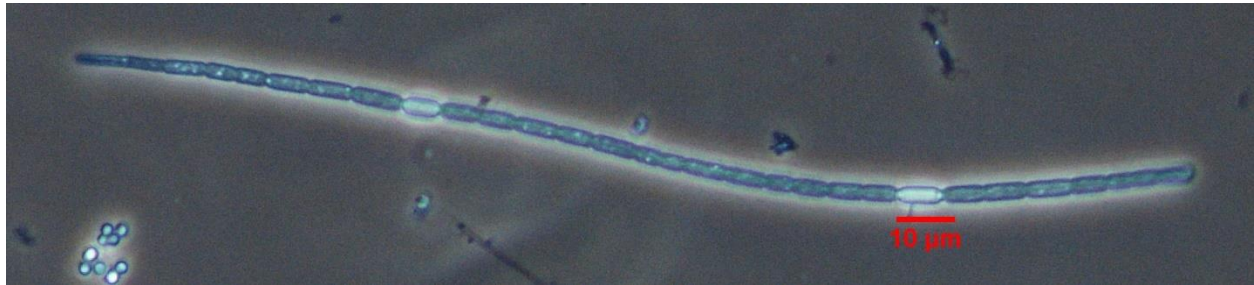
Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>
	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>



Recommendations

Based on these observations, analysis for microcystins is recommended for **all** samples. Additional screening for saxitoxins, anatoxin-a, and cylindrospermopsin is recommended for samples **2** and **9**.

**Micrographs**



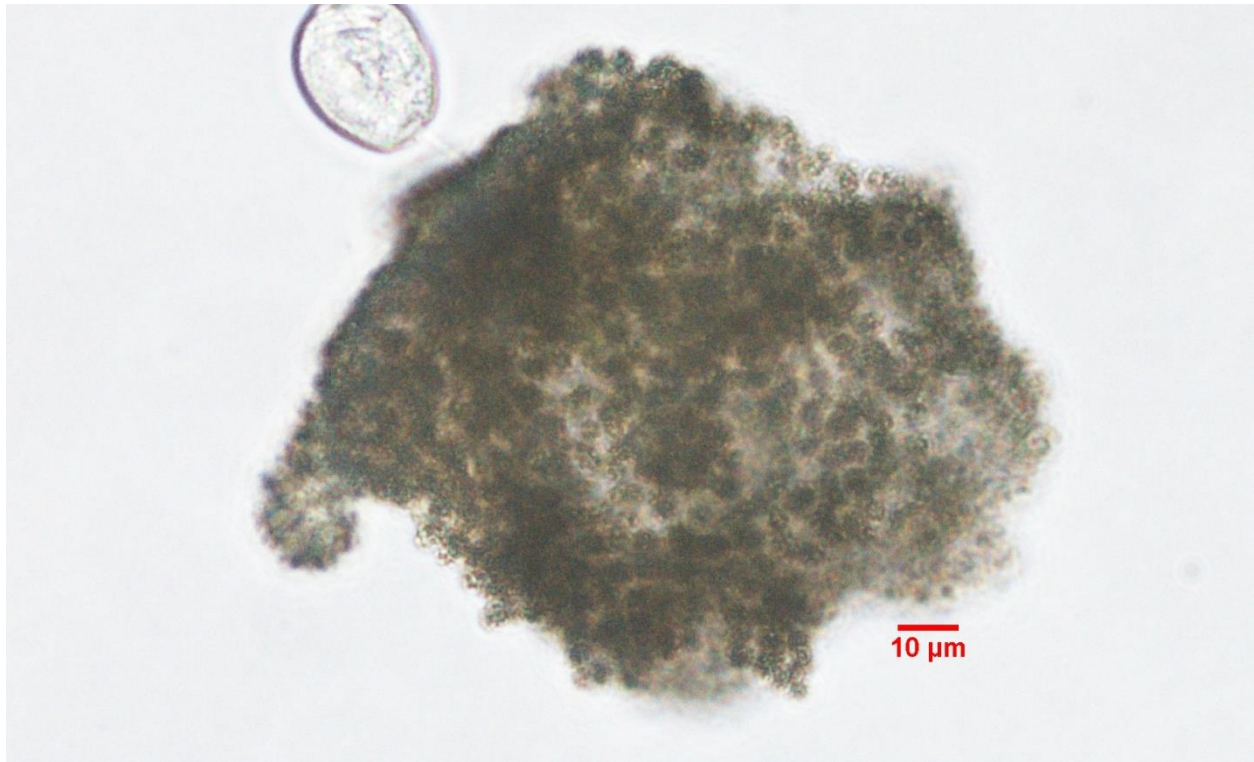
*Cuspidothrix* sp. at 400X (1)



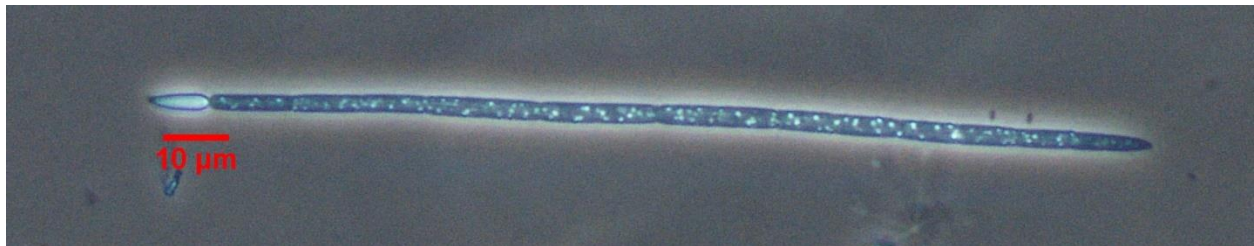
*Dolichospermum* sp. at 400X (1)



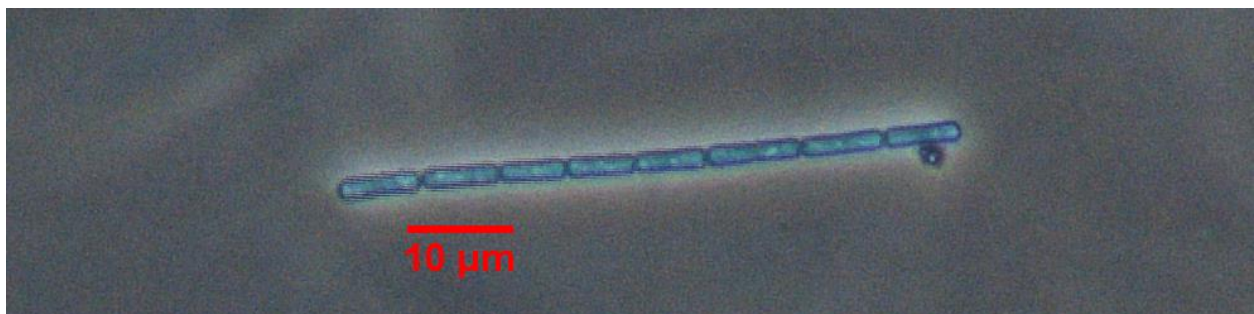
*Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. at 400X (2)



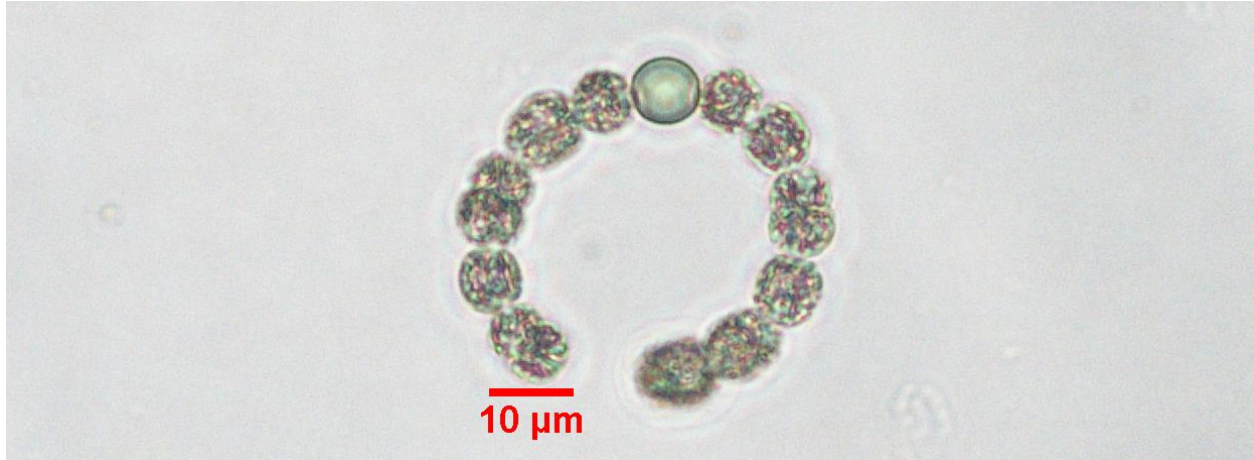
*Microcystis* sp. at 400X (1)



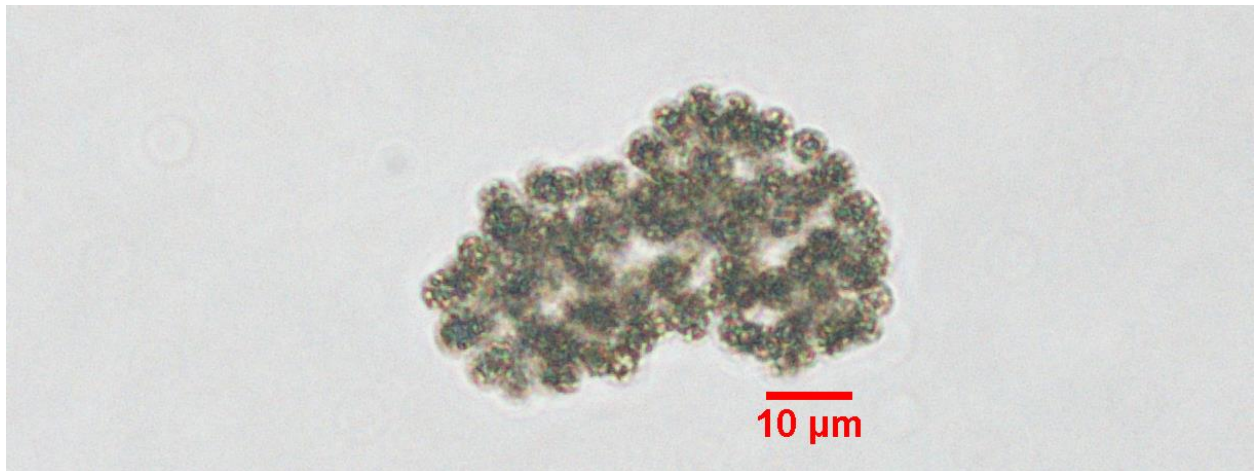
*Raphidiopsis* sp. at 400X (1)



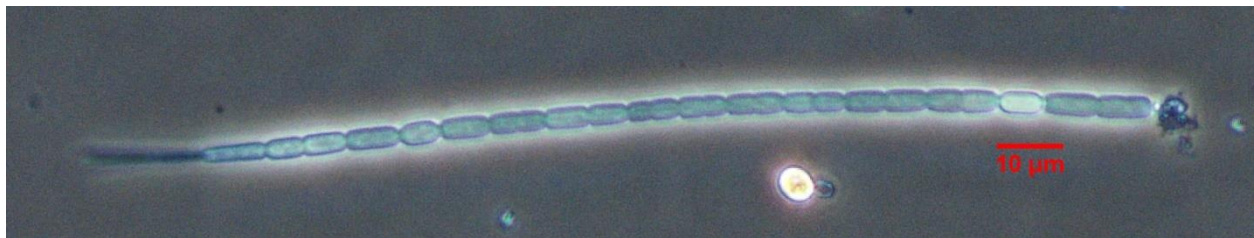
*Pseudanabaena* sp. at 400X (1)



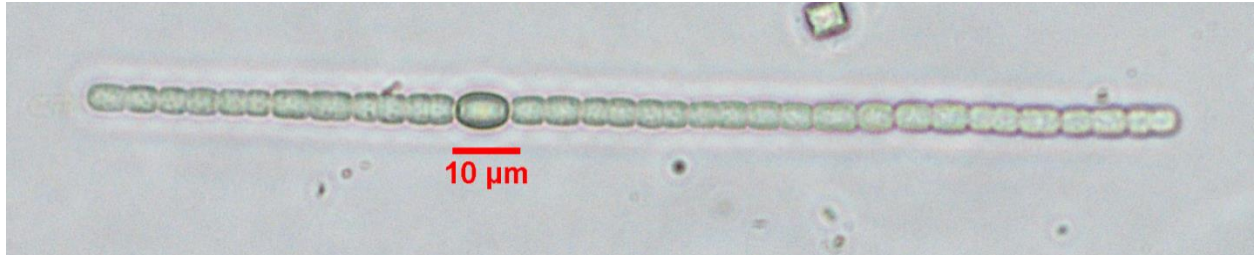
*Dolichospermum* sp. at 400X (9)



*Microcystis* sp. at 400X (9)



*Cuspidothrix* sp. at 400X (9)



cf. *Aphanizomenon* sp. at 400X (9)



*Pseudanabaena* sp. at 400X (9)

Submitted by:

*Amanda Foss*

Amanda Foss, M.S.

Date:

June 2, 2021

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### Microcystins/Nodularins & Saxitoxins Report

*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 4.6 °C upon arrival  
 Report# 210529\_BlueGreen  
 Date Prepared: 18 June 2021  
 Prepared by: Amanda Foss

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
1	W.P. Franklin	29 May 2021
2	W.P. Franklin	29 May 2021
3	W.P. Franklin	29 May 2021
4	W.P. Franklin	29 May 2021
5	W.P. Franklin	29 May 2021
6	W.P. Franklin	29 May 2021
7	W.P. Franklin	29 May 2021
8	W.P. Franklin	29 May 2021

**Toxins** –Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

#### Abbreviations

NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Regent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

**Quality Control**

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	5	LFSM	135% <sup>N</sup>
MC-LR (as MMPB)	2.0	8	LFSM	85%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	71 ± 13%
STX	0.2	1	LFSM	100%
STX	0.2	6	LFSM	103%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ±50%; IS ± 50%

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received

Table 3: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
1	STX	1	0.08	5.8%	0.08	<b>0.09</b>
		1	0.09		0.09	
1 LFSM	STX	1	0.29	2.5%	0.29	<b>0.29</b>
		1	0.28		0.28	
2	STX	1	0.05	38.4%	0.05	<b>0.07</b>
		1	0.08		0.08	
3	STX	1	0.09	0.9%	0.09	<b>0.09</b>
		1	0.09		0.09	
4	STX	1	0.10	1.5%	0.10	<b>0.10</b>
		1	0.10		0.10	
5	STX	1	0.10	3.4%	0.10	<b>0.10</b>
		1	0.09		0.09	
6	STX	1	0.07	0.6%	0.07	<b>0.07</b>
		1	0.07		0.07	
6 LFSM	STX	1	0.27	2.0%	0.27	<b>0.28</b>
		1	0.28		0.28	
7	STX	1	0.09	3.1%	0.09	<b>0.09</b>
		1	0.09		0.09	
8	STX	1	0.09	5.5%	0.09	<b>0.09</b>
		1	0.08		0.08	

Table 4: STX-ELISA Quality Control Value Table

Date Analyzed:	11 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.999	≥0.98	PASS
<b>%CV range STDs:</b>	0.3-5.0%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	78%	±40% True Value	PASS
<b>%CV range LFB:</b>	3.7%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	80%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS




**Summary of Results**

Table 6: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method and saxitoxins (STX). All data is reported as ng/mL (ppb).

<b>Sample</b>	<b>MCs/NODs (ng/mL)</b>	<b>STX (ng/mL)</b>
1	<b>7.94</b>	<b>0.09</b>
2	<b>3.59</b>	<b>0.07</b>
3	<b>0.99</b>	<b>0.09</b>
4	<b>1.01</b>	<b>0.10</b>
5	<b>1.54</b>	<b>0.10</b>
6	<b>310</b>	<b>0.07</b>
7	<b>1.55</b>	<b>0.09</b>
8	<b>1.46</b>	<b>0.09</b>
<i>MRL (ng/mL):</i>	<i>0.06</i>	<i>0.05</i>
<i>Analyst Initials:</i>	<i>AF</i>	<i>KC</i>
<i>Date Analyzed:</i>	<i>6/16/2021</i>	<i>6/11/2021</i>

**Interpretations:**

Microcystins/nodularins were above the current ‘EPA Recommended Value for Recreational Criteria and Swimming Advisory’ of 8.0 ng/mL (ppb) (EPA, 2019) in the 6 – W.P. Franklin sample. Saxitoxins were detected in all samples at sub-ppb levels using a saxitoxin specific ELISA.

Submitted by:   
 Mark T. Aubel, Ph.D.  
 Lab Director  
 Date: June 18, 2021

*The results in this report relate only to the samples listed above.  
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## Potentially Toxicogenic (PTOX) Cyanobacteria Report

*Project: BlueGreen Water Tech*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 4.6 °C upon arrival  
 Report# 210529\_PTOX\_BlueGreen  
 Date Prepared: 2 June 2021  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
1	W.P. Franklin	5/29/21
2	W.P. Franklin	5/29/21
3	W.P. Franklin	5/29/21
4	W.P. Franklin	5/29/21
5	W.P. Franklin	5/29/21
6	W.P. Franklin	5/29/21
7	W.P. Franklin	5/29/21
8	W.P. Franklin	5/29/21

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 1

The potentially toxicogenic (PTOX) cyanobacteria *Dolichospermum* sp. (>20 filaments per mL; mostly partial), *Raphidiopsis* sp. (>20 filaments per mL), *Cuspidothrix* sp. (>15 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (>10 colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**2**

The PTOX cyanobacteria *Dolichospermum* sp. (>30 filaments per mL; mostly partial), *Cuspidothrix* sp. (>15 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>15 filaments per mL), *Raphidiopsis* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq 4$  colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**3**

The PTOX cyanobacteria *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>25 filaments per mL), *Cuspidothrix* sp. (>25 filaments per mL), *Raphidiopsis* sp. (>15 filaments per mL), *Microcystis* sp. ( $\geq 5$  colonies per mL), *Dolichospermum* sp. ( $\geq 4$  partial filaments), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**4**

The PTOX cyanobacteria *Raphidiopsis* sp. (>30 filaments per mL; straight and coiled morphologies), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>25 filaments per mL), *Cuspidothrix* sp. (>15 filaments per mL), *Dolichospermum* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq 3$  colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**5**

The PTOX cyanobacteria *Cuspidothrix* sp. ( $\geq 25$  filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>20 filaments per mL), *Raphidiopsis* sp. (>15 filaments per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Microcystis* sp. ( $\geq 5$  colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**6**

The sample was dominated by the PTOX cyanobacterium *Microcystis* sp. (>200 colonies per mL). Other PTOX cyanobacteria observed included *Dolichospermum* sp. (>30 filaments per mL; mostly partial), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>15 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Raphidiopsis* sp. (>5 filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**7**

The PTOX cyanobacteria *Cuspidothrix* sp. (>30 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>25 filaments per mL), *Dolichospermum* sp. ( $\geq 20$  partial filaments per mL), *Raphidiopsis* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq 4$  colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

**8**

The PTOX cyanobacteria *Cuspidothrix* sp. (>30 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>20 filaments per mL), *Raphidiopsis* sp. (>15 filaments per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Microcystis* sp. (≥5 colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL) were observed.

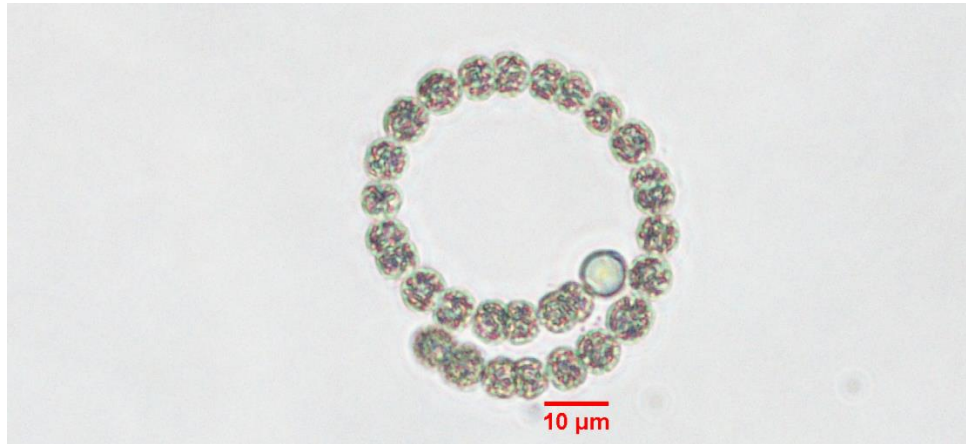
Potential toxin producing genera observed include:

Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>
	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>

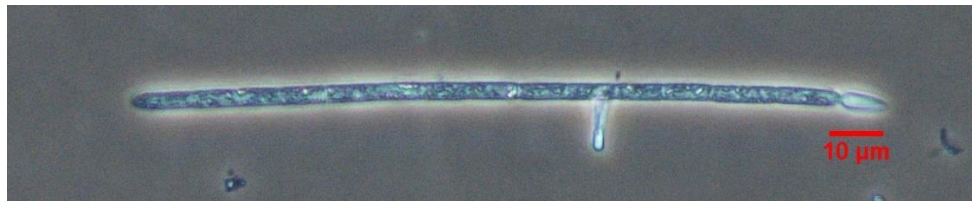
Recommendations

Based on the observed density of the PTOX cyanobacterium *Microcystis*, analysis for microcystins is recommended for sample **6**. Toxin recommendations for the other samples in this set are pending results from the previous sample collection (5/28/2021).

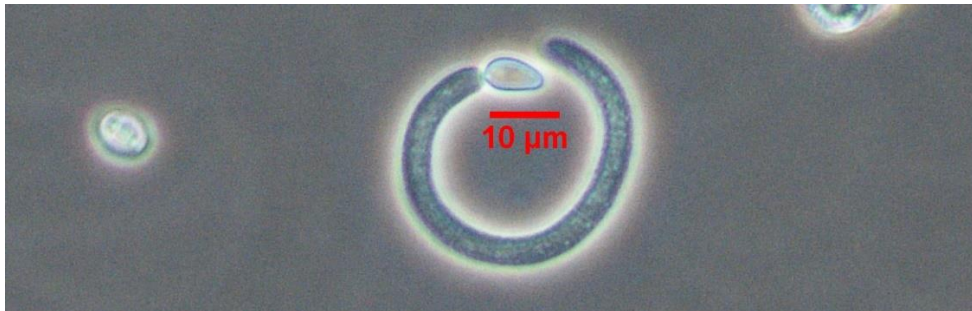
**Micrographs**



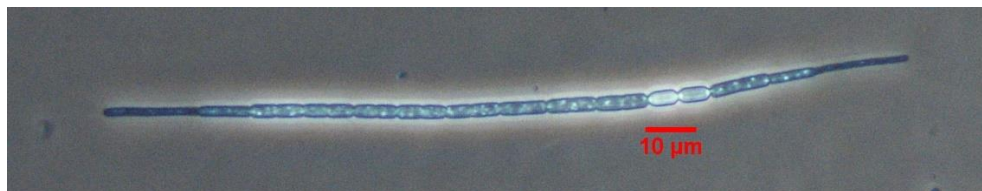
*Dolichospermum* sp. at 400X (1)



*Raphidiopsis* sp. (straight) at 400X (1)



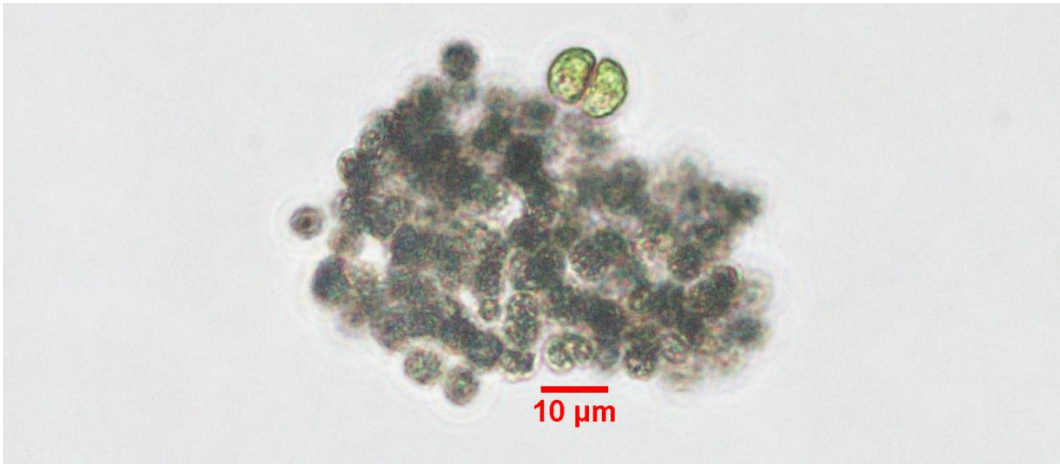
*Raphidiopsis* sp. (coiled) at 400X (4)



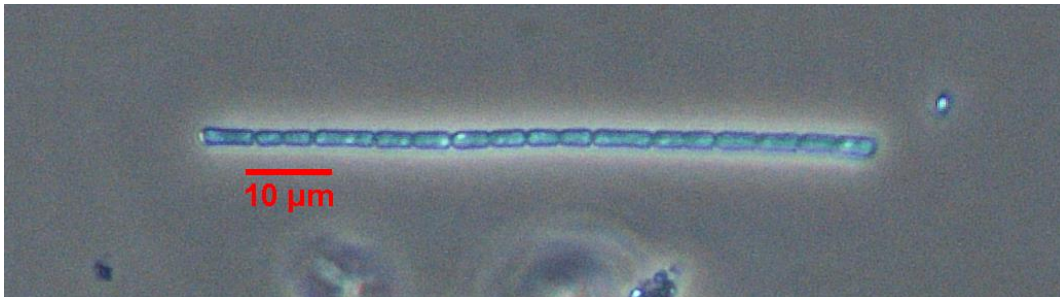
*Cuspidothrix* sp. at 400X (1)



*Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. at 400X (2)



*Microcystis* sp. at 400X (1)



*Pseudanabaena* sp. at 400X (1)

Submitted by:

*Amanda Foss*  
Amanda Foss, M.S.

Date:

June 2, 2021

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### Microcystins/Nodularins & Saxitoxins Report

*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 4.4 °C upon arrival  
 Report# 210530\_BlueGreen  
 Date Prepared: 18 June 2021  
 Prepared by: Amanda Foss

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
1	W.P. Franklin	30 May 2021
2	W.P. Franklin	30 May 2021
3	W.P. Franklin	30 May 2021
4	W.P. Franklin	30 May 2021
5	W.P. Franklin	30 May 2021
6	W.P. Franklin	30 May 2021
7	W.P. Franklin	30 May 2021
8	W.P. Franklin	30 May 2021

**Toxins** –Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

#### Abbreviations

NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Reagent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).



### Quality Control

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	2	LFSM	119%
MC-LR (as MMPB)	2.0	5	LFSM	106%
MC-LR (as MMPB)	2.0	7	LFSM	114%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	60 ± 18%
STX	0.2	1	LFSM	100%
STX	0.2	6	LFSM	103%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ± 50%; IS ± 50%

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received

Table 3: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
1	STX	1	0.11	23.2%	0.11	<b>0.10</b>
		1	0.08		0.08	
1 LFSM	STX	1	0.29	1.2%	0.29	<b>0.30</b>
		1	0.30		0.30	
2	STX	1	0.10	1.0%	0.10	<b>0.10</b>
		1	0.10		0.10	
3	STX	1	0.10	0.3%	0.10	<b>0.10</b>
		1	0.10		0.10	
4	STX	1	0.09	9.7%	0.09	<b>0.10</b>
		1	0.11		0.11	
5	STX	1	0.11	2.1%	0.11	<b>0.11</b>
		1	0.10		0.10	
6	STX	1	0.09	8.5%	0.09	<b>0.09</b>
		1	0.08		0.08	
6 LFSM	STX	1	0.27	8.5%	0.27	<b>0.29</b>
		1	0.31		0.31	
7	STX	1	0.08	3.8%	0.08	<b>0.08</b>
		1	0.08		0.08	
8	STX	1	0.07	11.5%	0.07	<b>0.08</b>
		1	0.08		0.08	

Table 4: STX-ELISA Quality Control Value Table

Date Analyzed:	15 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.998	≥0.98	PASS
<b>%CV range STDs:</b>	0.7-4.4%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	84%	±40% True Value	PASS
<b>%CV range LFB:</b>	7.1%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	70%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS

## Summary of Results

Table 6: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method and saxitoxins (STX). All data is reported as ng/mL (ppb).

Sample	MCs/NODs (ng/mL)	STX (ng/mL)
1	0.27	0.10
2	1.04	0.10
3	0.80	0.10
4	2.91	0.10
5	1.85	0.11
6	1.76	0.09
7	0.70	0.08
8	0.73	0.08
<hr/>		
MRL (ng/mL):	0.06	0.05
Analyst Initials:	AF	KC
Date Analyzed:	6/16/2021	6/15/2021

### Interpretations:

Microcystins/nodularins were below the current 'EPA Recommended Value for Recreational Criteria and Swimming Advisory' of 8.0 ng/mL (ppb) (EPA, 2019) in all samples. Saxitoxins were detected in all samples at sub-ppb levels using a saxitoxin specific ELISA.

Submitted by:



Mark T. Aubel, Ph.D.  
Lab Director

Date:

June 18, 2021

*The results in this report relate only to the samples listed above.  
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## Potentially Toxicogenic (PTOX) Cyanobacteria Report

Project: BlueGreen Water Tech

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 4.4 °C upon arrival  
 Report# 210531\_PTOX\_BlueGreen  
 Date Prepared: 3 June 2021  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
1	W.P. Franklin	5/30/21
2	W.P. Franklin	5/30/21
3	W.P. Franklin	5/30/21
4	W.P. Franklin	5/30/21
5	W.P. Franklin	5/30/21
6	W.P. Franklin	5/30/21
7	W.P. Franklin	5/30/21
8	W.P. Franklin	5/30/21

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 1

The potentially toxicogenic (PTOX) cyanobacteria *Raphidiopsis* sp. (>40 filaments per mL), *Cuspidothrix* sp. (>25 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>15 filaments per mL), *Microcystis* sp. (≥8 colonies per mL), *Dolichospermum* sp. (≥2 partial filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**2**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Cuspidothrix* sp. (>20 filaments per mL), *Microcystis* sp. (>10 colonies per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq$ 10 filaments per mL), *Dolichospermum* sp. ( $\geq$ 3 partial filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**3**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>15 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq$ 8 colonies per mL), *Dolichospermum* sp. ( $\geq$ 1 partial filament per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**4**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>15 filaments per mL), *Microcystis* sp. ( $\geq$ 9 colonies per mL), *Cuspidothrix* sp. (>5 filaments per mL), *Dolichospermum* sp. (>5 partial filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**5**

The PTOX cyanobacteria *Raphidiopsis* sp. (>30 filaments per mL), *Cuspidothrix* sp. (>20 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq$ 15 filaments per mL), *Microcystis* sp. ( $\geq$ 6 colonies per mL), *Dolichospermum* sp. (>5 partial filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**6**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>15 filaments per mL), *Microcystis* sp. ( $\geq$ 9 colonies per mL), *Cuspidothrix* sp. (>5 filaments per mL), *Dolichospermum* sp. ( $\geq$ 3 partial filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**7**

The PTOX cyanobacteria *Raphidiopsis* sp. (>15 filaments per mL), *Cuspidothrix* sp. ( $\geq$ 10 filaments per mL), *Microcystis* sp. ( $\geq$ 9 colonies per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>5 filaments per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

**8**

The PTOX cyanobacteria *Raphidiopsis* sp. (>10 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Dolichospermum* sp. (>10 partial filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>5 filaments per mL), *Microcystis* sp. (≥3 colonies per mL), and *Pseudanabaena* sp. (<10 filaments per mL) were observed.

Potential toxin producing genera observed include:

Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>
	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>

Recommendations

Based on similar PTOX cyanobacterial composition observed in previous samples, toxin recommendations are pending results from the 5/28/2021 collection.

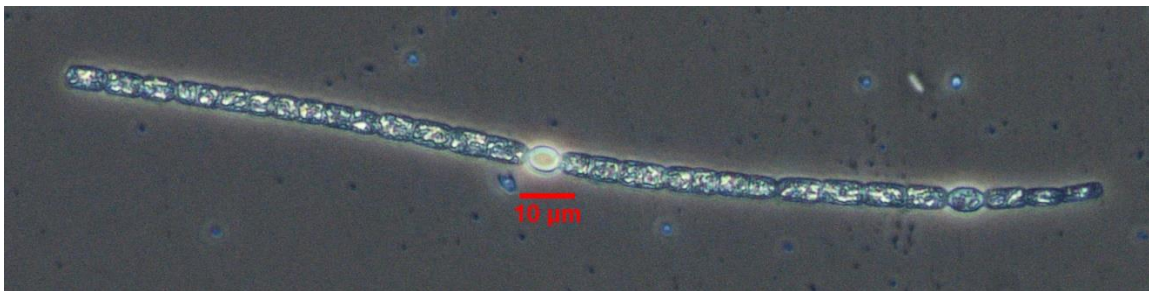
**Micrographs**



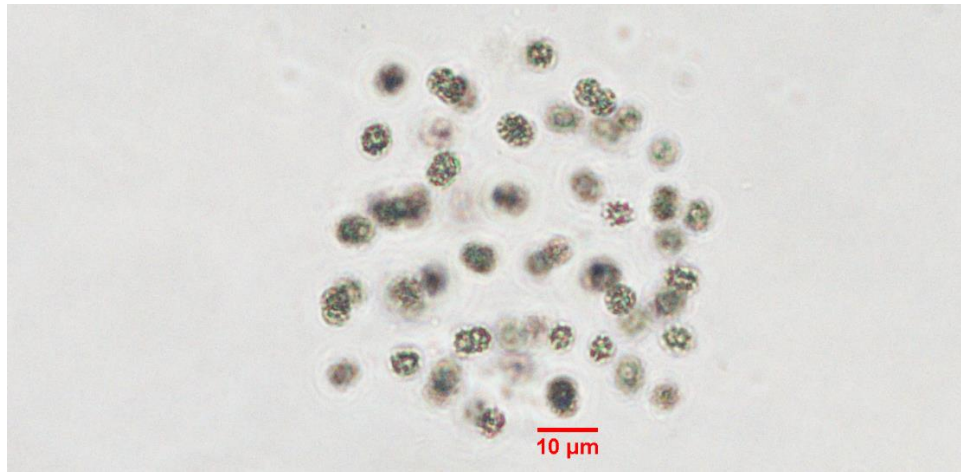
*Raphidiopsis* sp. at 400X (1)



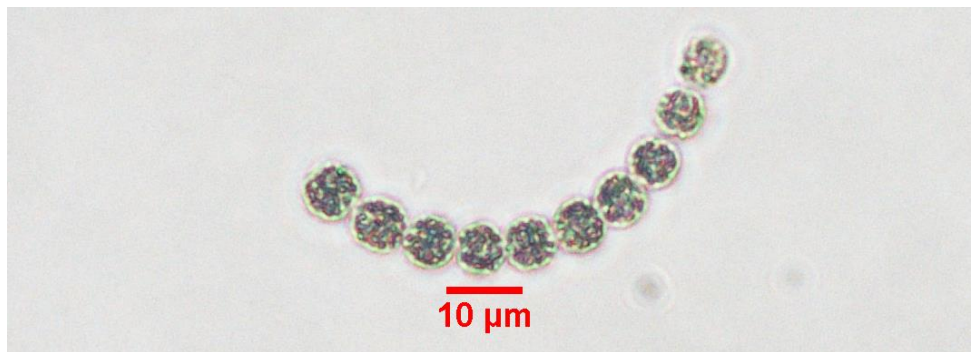
*Cuspidothrix* sp. at 400X (1)



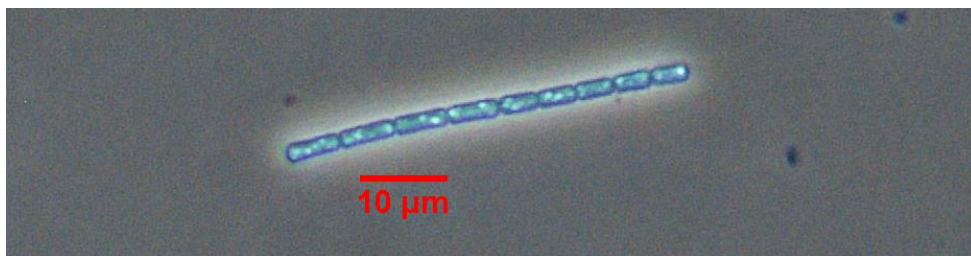
*Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. at 400X (4)



*Microcystis* sp. at 400X (1)



*Dolichospermum* sp. at 400X (1)



*Pseudanabaena* sp. at 400X (1)

Submitted by:

Amanda Foss  
Amanda Foss, M.S.

Date:

June 3, 2021

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### Microcystins/Nodularins & Saxitoxins Report

*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 2.6 °C upon arrival  
 Report# 210531\_BlueGreen  
 Date Prepared: 21 June 2021  
 Prepared by: Kamil Cieslik

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
1	W.P. Franklin	31 May 2021
2	W.P. Franklin	31 May 2021
3	W.P. Franklin	31 May 2021
4	W.P. Franklin	31 May 2021
5	W.P. Franklin	31 May 2021
6	W.P. Franklin	31 May 2021
7	W.P. Franklin	31 May 2021
8	W.P. Franklin	31 May 2021

**Toxins** –Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

#### Abbreviations

NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Regent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

### Quality Control

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	1	LFSM	101%
MC-LR (as MMPB)	2.0	6	LFSM	99%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	55 ± 78%
STX	0.2	1	LFSM	93%
STX	0.2	6	LFSM	88%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ± 50%; IS ± 50%

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received

Table 3: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
1	STX	1	0.10	0.4%	0.10	<b>0.11</b>
		1	0.11		0.11	
1 LFSM	STX	1	0.28	3.9%	0.28	<b>0.29</b>
		1	0.30		0.30	
2	STX	1	0.10	0.2%	0.10	<b>0.10</b>
		1	0.10		0.10	
3	STX	1	0.08	12.7%	0.08	<b>0.09</b>
		1	0.09		0.09	
4	STX	1	0.10	0.9%	0.10	<b>0.10</b>
		1	0.09		0.09	
5	STX	1	0.09	0.8%	0.09	<b>0.09</b>
		1	0.09		0.09	
6	STX	1	0.10	6.1%	0.10	<b>0.10</b>
		1	0.09		0.09	
6 LFSM	STX	1	0.27	0.7%	0.27	<b>0.27</b>
		1	0.27		0.27	
7	STX	1	0.10	7.6%	0.10	<b>0.10</b>
		1	0.09		0.09	
8	STX	1	0.08	2.1%	0.08	<b>0.08</b>
		1	0.08		0.08	

Table 4: STX-ELISA Quality Control Value Table

Date Analyzed:	17 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.998	≥0.98	PASS
<b>%CV range STDs:</b>	0.7-4.2%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	80%	±40% True Value	PASS
<b>%CV range LFB:</b>	0.0%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	80%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS


**Summary of Results**

Table 6: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method and saxitoxins (STX). All data is reported as ng/mL (ppb).

<b>Sample</b>	<b>MCs/NODs (ng/mL)</b>	<b>STX (ng/mL)</b>
1	<b>0.48</b>	<b>0.11</b>
2	<b>2.02</b>	<b>0.10</b>
3	<b>0.99</b>	<b>0.09</b>
4	<b>0.7</b>	<b>0.10</b>
5	<b>0.54</b>	<b>0.09</b>
6	<b>0.53</b>	<b>0.10</b>
7	<b>0.67</b>	<b>0.10</b>
8	<b>4.08</b>	<b>0.08</b>
<i>MRL (ng/mL):</i>	<i>0.06</i>	<i>0.05</i>
<i>Analyst Initials:</i>	<i>AF</i>	<i>KC</i>
<i>Date Analyzed:</i>	<i>6/18/2021</i>	<i>6/17/2021</i>

**Interpretations:**

Microcystins/nodularins were below the current ‘EPA Recommended Value for Recreational Criteria and Swimming Advisory’ of 8.0 ng/mL (ppb) (EPA, 2019) in all samples. Saxitoxins were detected in all samples at sub-ppb levels using a saxitoxin specific ELISA.

Submitted by:   
Mark T. Aubel, Ph.D.  
Lab Director

Date: June 21, 2021

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## Potentially Toxicogenic (PTOX) Cyanobacteria Report

*Project: BlueGreen Water Tech*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 2 June 2021  
 Sample Condition: 2.6 °C upon arrival  
 Report# 210531\_PTOX\_BlueGreen  
 Date Prepared: 3 June 2021  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
1	W.P. Franklin	5/31/21
2	W.P. Franklin	5/31/21
3	W.P. Franklin	5/31/21
4	W.P. Franklin	5/31/21
5	W.P. Franklin	5/31/21
6	W.P. Franklin	5/31/21
7	W.P. Franklin	5/31/21
8	W.P. Franklin	5/31/21

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 1

The potentially toxicogenic (PTOX) cyanobacteria *Raphidiopsis* sp. (>35 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (≥8 colonies per mL), *Dolichospermum* sp. (≥5 partial filaments per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**2**

The PTOX cyanobacteria *Raphidiopsis* sp. (>40 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq$ 15 filaments per mL), *Dolichospermum* sp. (>10 partial filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq$ 6 colonies per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**3**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>20 filaments per mL), *Cuspidothrix* sp. (>20 filaments per mL), *Dolichospermum* sp. (>10 partial filaments per mL), *Microcystis* sp. ( $\geq$ 4 colonies per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**4**

The PTOX cyanobacteria *Raphidiopsis* sp. (>20 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. ( $\geq$ 6 colonies per mL), *Dolichospermum* sp. (>5 partial filaments per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**5**

The PTOX cyanobacteria *Raphidiopsis* sp. (>15 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>15 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Dolichospermum* sp. (>10 partial filaments per mL), *Microcystis* sp. ( $\geq$ 7 colonies per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**6**

The PTOX cyanobacteria *Raphidiopsis* sp. (>20 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>5 filaments per mL), *Dolichospermum* sp. ( $\geq$ 3 partial filaments per mL), *Microcystis* sp. ( $\geq$ 3 colonies per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**7**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Microcystis* sp. ( $\geq$ 4 colonies per mL), *Planktothrix* sp. (1 filament per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

**8**

The PTOX cyanobacteria *Raphidiopsis* sp. (>25 filaments per mL), *Cuspidothrix* sp. (>15 filaments per mL), *Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. (>15 filaments per mL), *Microcystis* sp. (>10 colonies per mL), *Dolichospermum* sp. (>5 partial filaments per mL), and *Pseudanabaena* sp. (<10 small filaments per mL) were observed.

Potential toxin producing genera observed include:

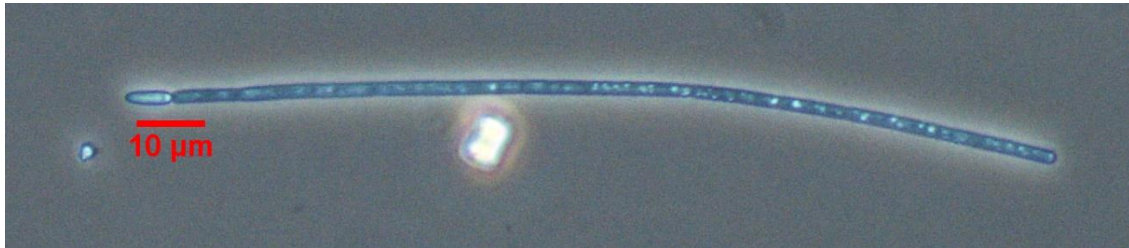
Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysochlorum</i>	<i>Chrysochlorum</i>	<i>Chrysochlorum</i>
<i>Planktothrix</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>
	<i>Planktothrix</i>	<i>Planktothrix</i>	

Recommendations

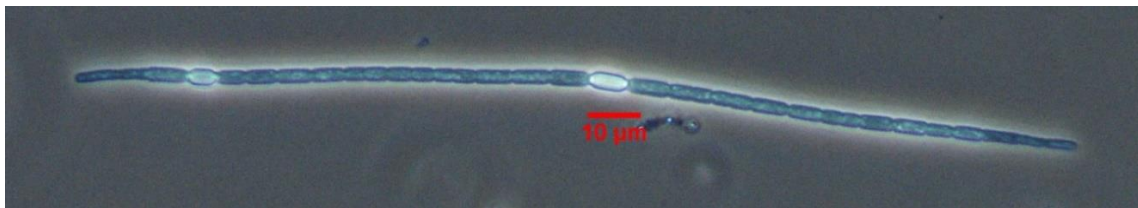
Based on similar PTOX cyanobacterial composition observed in previous samples, toxin recommendations are pending results from the 5/28/2021 collection.



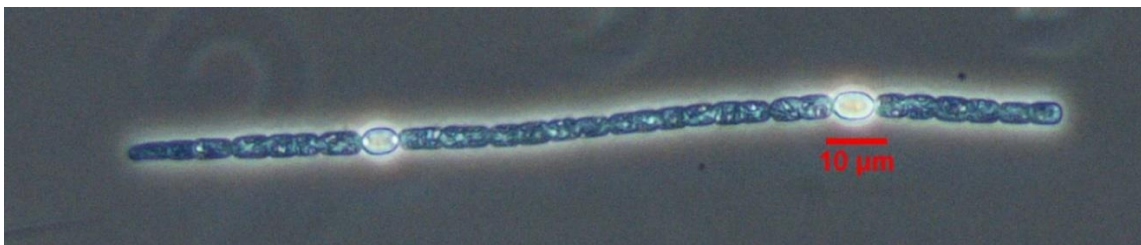
**Micrographs**



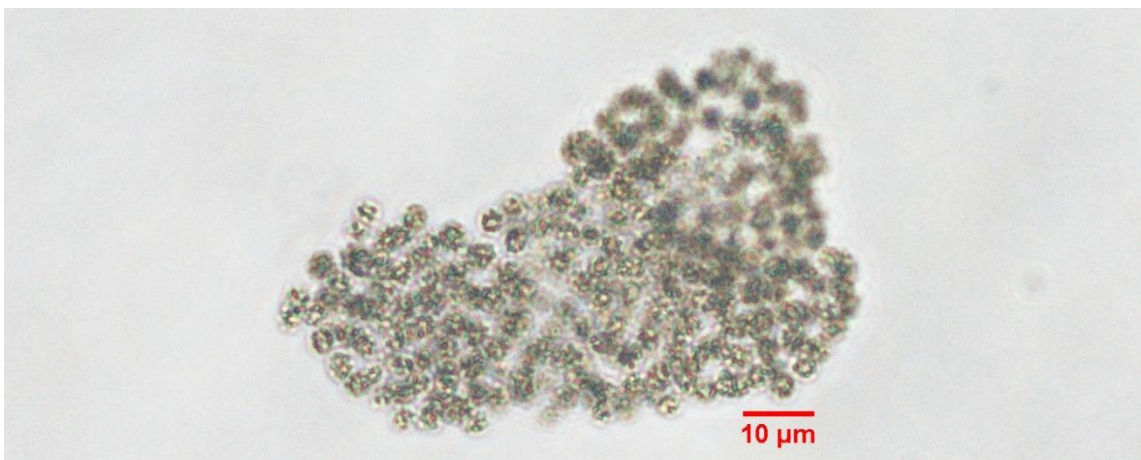
*Raphidiopsis* sp. (straight) at 400X (1)



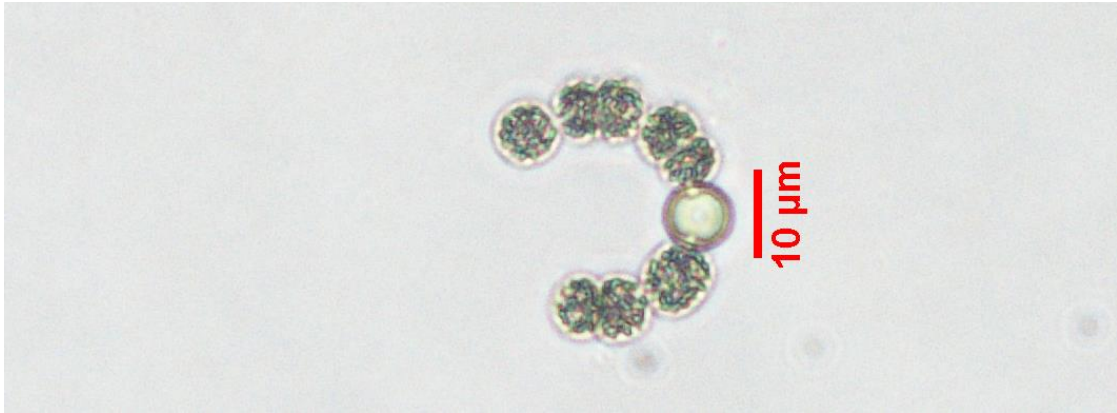
*Cuspidothrix* sp. at 400X (1)



*Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. at 400X (1)



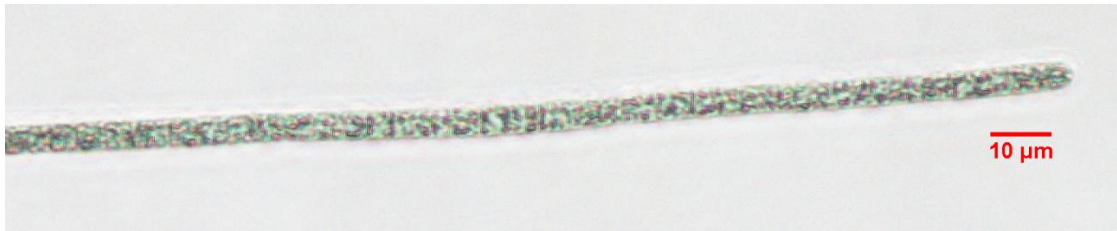
*Microcystis* sp. at 400X (1)



*Dolichospermum* sp. at 400X (1)



*Pseudanabaena* sp. at 400X (4)



*Planktothrix* sp. at 400X (7)

Submitted by:

*Amanda Foss*

Amanda Foss, M.S.

Date:

June 3, 2021

*The results in this report relate only to the samples listed above.  
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### Microcystins/Nodularins & Saxitoxins Report

*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 10 June 2021  
 Sample Condition: 5.9 °C upon arrival  
 Report# 210602-06\_BlueGreen  
 Date Prepared: 23 June 2021  
 Prepared by: Kamil Cieslik

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
5	W.P. Franklin	2 June 2021
9	W.P. Franklin	2 June 2021
10	W.P. Franklin	2 June 2021
12	W.P. Franklin	2 June 2021
K20	C43	6 June 2021
K21	C43	6 June 2021

**Toxins** –Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

<b>Abbreviations</b>			
NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Regent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

## Quality Control

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	12	LFSM	76%
MC-LR (as MMPB)	2.0	K21	LFSM	107%
MC-LR (as MMPB)	10	K21	LFSM	119%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	74 ± 12%
STX	0.2	5	LFSM	105%
STX	0.2	K21	LFSM	95%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ± 50%; IS ± 50%

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received

Table 3: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
5	STX	1	0.08	2.6%	0.08	<b>0.09</b>
		1	0.09		0.09	
5 LFSM	STX	1	0.30	1.5%	0.30	<b>0.30</b>
		1	0.29		0.29	
9	STX	1	0.05	17.9%	0.05	<b>0.06</b>
		1	0.07		0.07	
10	STX	1	0.09	3.2%	0.09	<b>0.09</b>
		1	0.09		0.09	
12	STX	1	0.08	0.5%	0.08	<b>0.08</b>
		1	0.08		0.08	
K20	STX	1	0.05	2.8%	0.05	<b>0.05</b>
		1	0.05		0.05	
K21	STX	1	0.02	29.2%	<0.05	<b>ND</b>
		1	0.02		<0.05	
K21 LFSM	STX	1	0.20	5.4%	0.20	<b>0.21</b>
		1	0.22		0.22	

Table 4: STX-ELISA Quality Control Value Table

Date Analyzed:	23 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.998	≥0.98	PASS
<b>%CV range STDs:</b>	0.1-8.5%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	83%	±40% True Value	PASS
<b>%CV range LFB:</b>	4.7%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	60%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS

## Summary of Results

Table 5: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method and saxitoxins (STX). All data is reported as ng/mL (ppb).

Sample	MCs/NODs (ng/mL)	STX (ng/mL)
5	0.60	0.09
9	0.58	0.06
10	0.36	0.09
12	0.88 ± 0.11	0.08
K20	942 ± 81	0.05
K21	2.76	ND
<hr/>		
MRL (ng/mL):	0.06	0.05
Analyst Initials:	AF	KC
Date Analyzed:	6/18/2021 6/21/2021	6/23/2021

### Interpretations:

The levels of Adda MCs/NODs detected in the K20 sample (**942 ppb**) exceeds the current 'Draft EPA Recommended Value for Recreational Criteria and Swimming Advisory', which is currently 8 ng/mL (ppb) total microcystins. The WHO recreational guidance value for microcystin is currently 24 ng/mL (ppb) (World Health Organization (WHO), 2020a).

Saxitoxins were detected in all samples (**K21 sample exception**) at sub-ppb levels using a saxitoxin specific ELISA.

World Health Organization (WHO), 2020a. Cyanobacterial toxins: microcystins. Guidel. Drink. Qual. Guidel. Safe Recreat. Water Environ. 63.

Submitted by:



Mark T. Aubel, Ph.D.  
Lab Director

Date:

June 23, 2021

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## Potentially Toxicogenic (PTOX) Cyanobacteria Report

*Project: BlueGreen Water Tech*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 10 June 2021  
 Sample Condition: 5.9 °C upon arrival  
 Report# 210602-210606\_PTOX\_BlueGreen  
 Date Prepared: 14 June 2021  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
5	W.P. Franklin	6/2/21*
9	W.P. Franklin	6/2/21*
10	W.P. Franklin	6/2/21*
12	W.P. Franklin	6/2/21*
K20	C43	6/6/21*
K21	C43	6/6/21*

*\*The sample collection date for the W.P. Franklin site and the sample analysis date for the C43 sample set was outside the holding time for unpreserved algal identification samples (<5 days). This may have impacted the results.*

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 5

The potentially toxicogenic (PTOX) cyanobacteria observed included *Microcystis* sp. ( $\geq 30$  colonies per mL), *Dolichospermum* sp. ( $> 25$  filaments per mL; mostly partial), *Cuspidothrix* sp. ( $> 25$  filaments per mL), *Raphidiopsis raciborskii* ( $> 10$  filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 10$  filaments per mL), and *Pseudanabaena* sp. ( $< 100$  filaments per mL).



## 9

The PTOX cyanobacteria observed included *Microcystis* sp. ( $\geq 6$  colonies per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 5$  filaments per mL), *Dolichospermum* sp. ( $\geq 4$  filaments per mL; mostly partial), and *Pseudanabaena* sp. ( $< 100$  filaments per mL).

## 10

The PTOX cyanobacteria observed included *Microcystis* sp. ( $\geq 15$  colonies per mL), *Raphidiopsis raciborskii* ( $> 10$  filaments per mL), *Cuspidothrix* sp. ( $> 5$  filaments per mL), *Dolichospermum* sp. ( $> 5$  filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 5$  filaments per mL), and *Pseudanabaena* sp. ( $< 100$  filaments per mL).

## 12

The PTOX cyanobacteria observed included *Microcystis* sp. ( $\geq 20$  colonies per mL), *Dolichospermum* sp. ( $> 5$  filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 4$  filaments per mL), *Cuspidothrix* sp. ( $\geq 2$  filaments per mL), and *Pseudanabaena* sp. ( $< 100$  filaments per mL).

## K20

The sample was dominated by the PTOX cyanobacteria *Microcystis* spp. Other PTOX cyanobacteria observed included *Dolichospermum* sp. ( $> 50$  filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 3$  filaments per mL), and *Cuspidothrix* sp. ( $\geq 1$  filament per mL).

## K21

The PTOX cyanobacteria observed included *Microcystis* spp. ( $\geq 20$  colonies per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 3$  filaments per mL), *Raphidiopsis raciborskii* ( $\geq 2$  filaments per mL), *Cuspidothrix* sp. ( $\geq 2$  filaments per mL), and *Pseudanabaena* sp. ( $< 10$  filaments per mL).

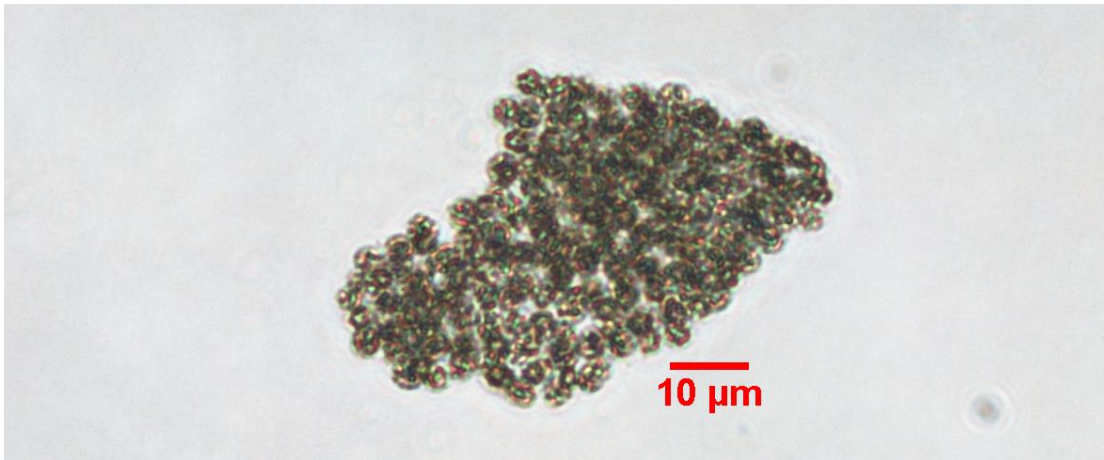
Potential toxin producing genera observed include:

Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>	<i>Chrysoosporum</i>
	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>

Recommendations

Based on these observations and previous toxin data, analysis for microcystins and saxitoxins are recommended for **all** samples.

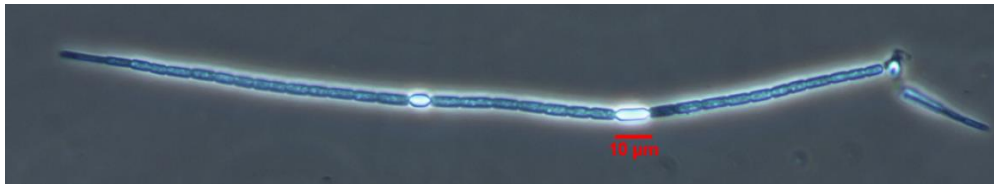
**Micrographs**



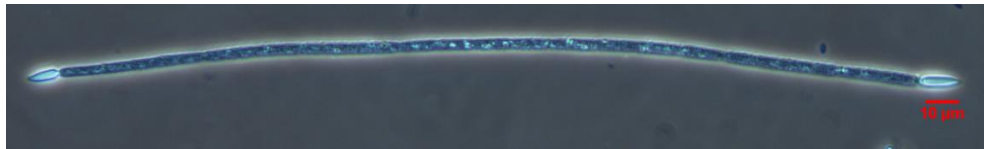
*Microcystis* sp. at 400X (5)



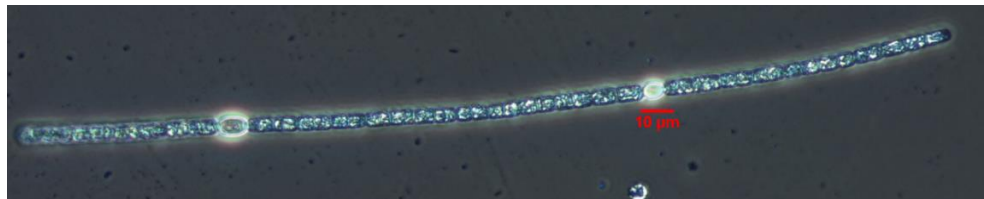
*Dolichopsermum* sp. at 400X (5)



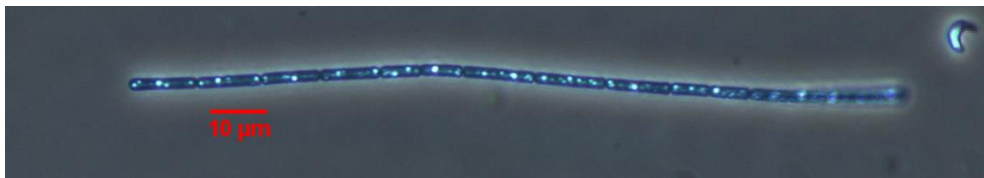
*Cuspidothrix* sp. at 400X (5)



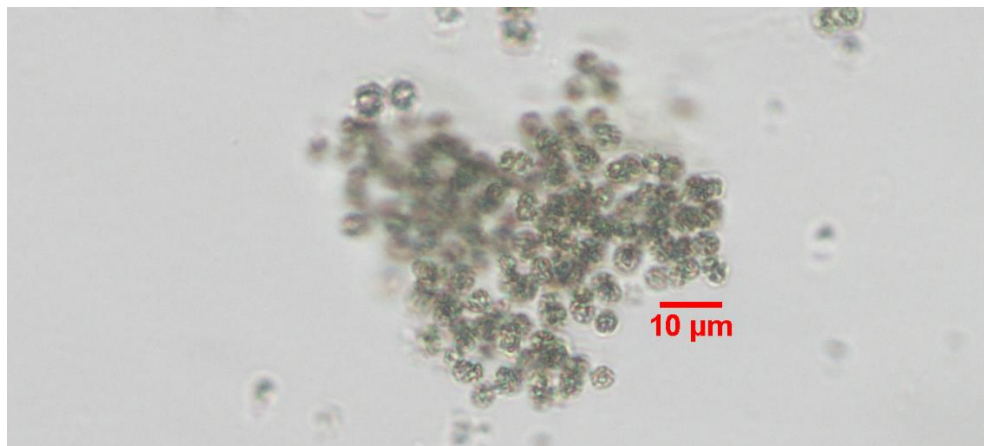
*Raphidiopsis raciborskii* at 400X (5)



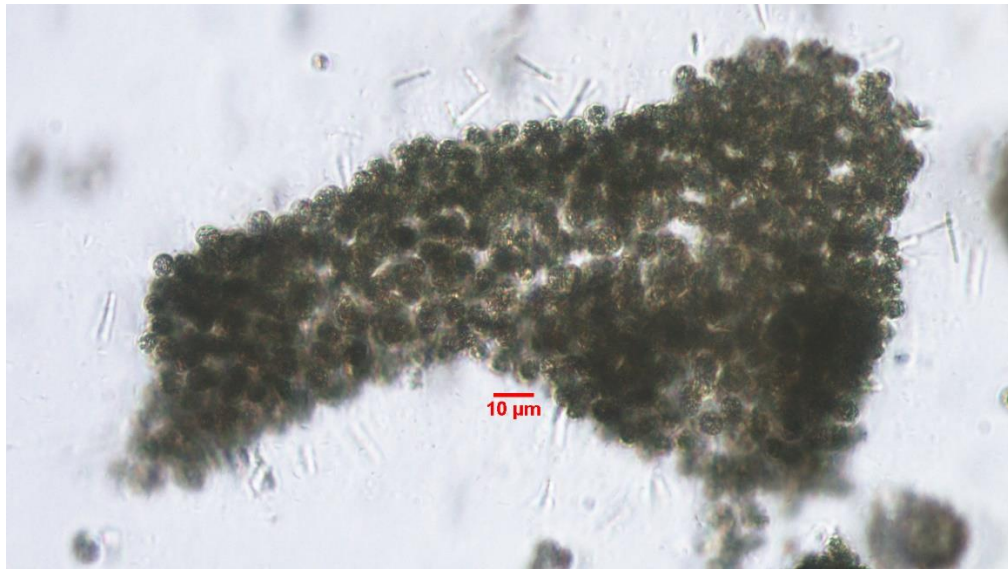
*Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. at 400X (5)



*Pseudanabaena* sp. at 400X (9)



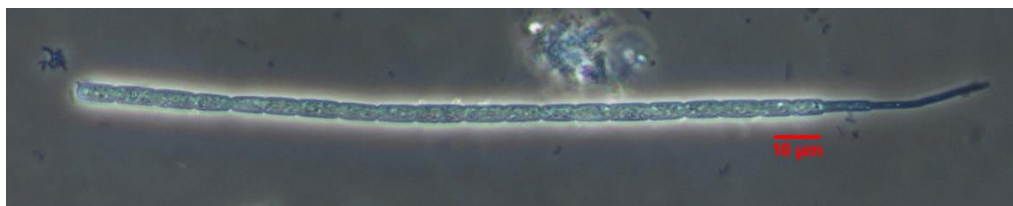
*Microcystis* sp. at 400X (K20)



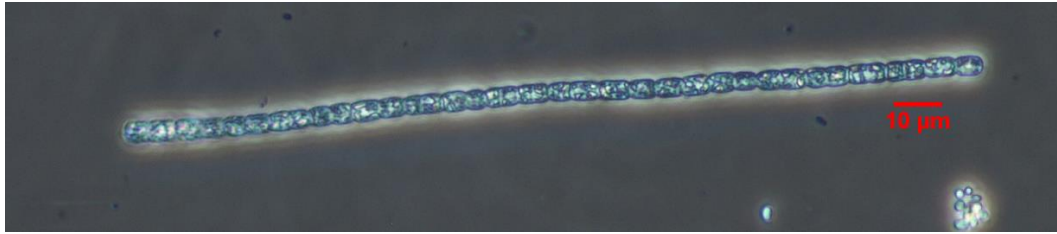
*Microcystis* sp. at 400X (K20)



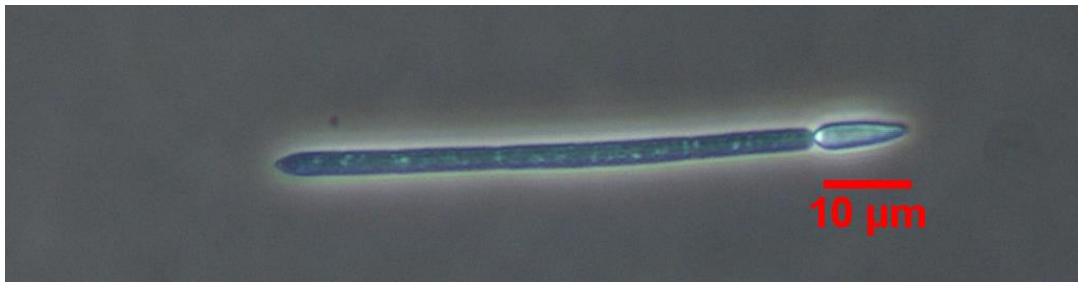
*Dolichospermum* sp. at 400X (K20)



*Cuspidothrix* sp. at 400X (K20)



*Aphanizomenon/Chrysoosporum/Sphaeropsermopsis* sp. at 400X (K21)



*Raphidiopsis raciborskii* at 400X (K21)



*Pseudanabaena* sp. at 400X (K21)

Submitted by:

*Amanda Foss*

Amanda Foss, M.S.

Date:

June 14, 2021

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## Microcystins/Nodularins & Saxitoxins Report

*Project: BlueGreen US Water Technologies Inc.*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies Inc.  
 Address: 1302 Waugh Drive, Ste. 482, Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
 Sample Receipt Date: 10 June 2021  
 Sample Condition: 4.6 °C upon arrival  
 Report# 210606\_BlueGreen  
 Date Prepared: 23 June 2021  
 Prepared by: Kamil Cieslik

Table 1: Samples analyzed and collection dates

<u>Sample ID</u>	<u>Description</u>	<u>Collection Date</u>
31	C-43	6 June 2021
32	C-43	6 June 2021
33	C-43	6 June 2021
34	C-43	6 June 2021
35	C-43	6 June 2021
36	C-43	6 June 2021
37	C-43	6 June 2021

**Toxins** –Adda Microcystins/Nodularins (MCs/NODs), 2-methyl-3-methoxy-4-phenylbutyric acid (MMPB), Saxitoxin (STX/PSTs)

Abbreviations			
NA	Not Applicable	LFSM	Lab Fortified Sample Matrix
MDL	Method Detection Limit	LFSMD	Lab Fortified Sample Matrix Duplicate
MQL	Method Quantification Limit	LD	Lab Duplicate
ND	Not Detected above the MDL	IS	Internal Standard
Blank	Regent Water free from interferences	—	Not Analyzed
LFB	Lab Fortified Blank	MRL	Method Reporting Limit
CCC	Continued Calibration Check	CV	Low-range calibration verification

## Sample Preparation

The samples were received and inverted for 60 seconds to mix. Subsets from each sample were removed prior to cell lysis for algal identification and enumeration purposes.

### *STX*

Subsets (100 mL) were sonicated to release toxins. Aliquots were prepared with IS and LFSMs (Table 2) and filtered (0.2  $\mu$ m PVDF) prior to analyses.

### *MCs/NODs by MMPB*

Aliquots (2 mL) spiked with IS ( $d_3$ -MMPB) were oxidized by the addition of 1 mL of a solution containing 0.1 M  $K_2CO_3$ , 0.05 M  $KMnO_4$  and 0.05 M  $NaIO_4$  for 1 hour, stopped with the addition of 40% sodium bisulfite and cleaned using 100 mg Strata X solid phase extraction (SPE). The extracts were reconstituted in DI, filtered (0.2  $\mu$ m PVDF), and analyzed.

## Analytical Techniques

### *Liquid chromatography mass spectrometry/mass spectrometry (LC-MS/MS)*

#### *MMPB*

The  $[M-H]^-$  ion of MMPB ( $m/z$  207) was fragmented and the product ion ( $m/z$  131) was monitored. The IS ( $d_3$ -MMPB) was also fragmented and monitored ( $m/z$  210 $\rightarrow$ 131). The internal standard method was implemented using a standard curve (0.25 – 100 ng/mL of oxidized MC-LR) to calculate LFSM returns.

### *Enzyme-Linked Immunosorbent Assay (ELISA)*

#### *STX*

A saxitoxin specific ELISA (Abraxis PN 52255B) was utilized for the detection and quantification of saxitoxin and related analogs (paralytic shellfish toxins – PSTs). The current method reporting limit is 0.05 ng/mL (ppb) based on kit sensitivity and dilution factors. Based on manufacture instructions, the STX ELISA is less cross-reactive to other PSTs and will likely underestimate total PSTs/Saxitoxins. Reported cross-reactivities are as follows: NEO (1.3%), dcSTX (29%), GTX2/3 (23%), GTX5 (23%), dcGTX2/3 (1.4%), dcNEO (0.6%) & GTX1/4 (<0.2%).

### Quality Control

Table 2: Quality Assurance/Quality Control (QA/QC) samples (IS and LFSM) prepared for analyses pre-extraction. Additional QA/QC checks included LFBs, continued calibration checks and external curves.

Analyte	Concentration (ng/mL)	Sample ID	QC Type	Return
MC-LR (as MMPB)	2.0	32	LFSM	85%
MC-LR (as MMPB)	2.0	36	LFSM	100%
MC-LR (as MMPB)	10	36	LFSM	83%
<i>d</i> <sub>3</sub> -MMPB	1.0	all aliquots	IS	63 ± 14%
STX	0.2	31	LFSM	95%
STX	0.2	36	LFSM	105%

\*Control limits: water LFSM ± 30%; complicated matrix LFSM and when LFSM within 2x MDL ± 50%; IS ± 50%

Qualifier	Flag
CL	Analytical result is estimated due to ineffective quenching.
J	Analyte was positively identified; the associated numerical value is estimated.
PT	The reported result is estimated because the sample was not analyzed within required holding time.
B	Analytical result is estimated. Analyte was detected in associated reagent blank as well as the samples.
E	Analytical result is estimated. Values achieved were outside calibration range.
N	Spiked sample control was outside limits
T	The reported result is estimated because the sample exceeded temperature threshold when received



Table 3: Raw ELISA Data

Sample ID	Analyte	Dilution Factor	Assay Values (ng/mL)	CV	Concentration (ng/mL)	Average (ng/mL)
31	STX	1	0.10	5.2%	0.10	<b>0.10</b>
		1	0.10		0.10	
31 LFSM	STX	1	0.29	0.3%	0.29	<b>0.29</b>
		1	0.29		0.29	
32	STX	1	0.04	19.1%	<0.05	<b>0.05<sup>J</sup></b>
		1	0.05		0.05	
33	STX	1	0.05	4.1%	0.05	<b>0.05</b>
		1	0.05		0.05	
34	STX	1	0.05	2.1%	0.05	<b>0.05</b>
		1	0.05		0.05	
35	STX	1	0.04	12.2%	<0.05	<b>ND</b>
		1	0.04		<0.05	
36	STX	1	0.06	3.5%	0.06	<b>0.06</b>
		1	0.06		0.06	
36 LFSM	STX	1	0.27	0.1%	0.27	<b>0.27</b>
		1	0.27		0.27	
37	STX	1	0.07	7.5%	0.07	<b>0.08</b>
		1	0.08		0.08	

Table 4: STX-ELISA Quality Control Value Table

Date Analyzed:	23 June 2021	Requirement	Pass/Fail
<b>R<sup>2</sup> value:</b>	0.998	≥0.98	PASS
<b>%CV range STDs:</b>	0.1-8.5%	≤15%	PASS
<b>LFB (0.2 ppb) recovery:</b>	83%	±40% True Value	PASS
<b>%CV range LFB:</b>	4.7%	<20%	PASS
<b>Low CCC (0.05 ppb) recovery:</b>	60%	±50% True Value	PASS
<b>LRB</b>	<0.03	<0.03	PASS

## Summary of Results

Table 5: Summary of results for total microcystins/nodularins (MCs/NODs) as measured through the MMPB method and saxitoxins (STX). All data is reported as ng/mL (ppb).


Sample	MCs/NODs (ng/mL)	STX (ng/mL)
31	1.26	0.10
32	1.59	0.05 <sup>J</sup>
33	500 ± 56	0.05
34	8.07	0.05
35	6.40	ND
36	1.74	0.06
37	1.28	0.08
<hr/>		
MRL (ng/mL):	0.06	0.05
Analyst Initials:	AF	KC
Date Analyzed:	6/18/2021 6/21/2021	6/23/2021

### Interpretations:

The levels of Adda MCs/NODs detected in the 33 sample (**500 ppb**) exceeds the current ‘Draft EPA Recommended Value for Recreational Criteria and Swimming Advisory’, which is currently 8 ng/mL (ppb) total microcystins. The WHO recreational guidance value for microcystin is currently 24 ng/mL (ppb) (World Health Organization (WHO), 2020a).

Saxitoxins were detected in all samples (**35 sample exception**) at sub-ppb levels using a saxitoxin specific ELISA.

World Health Organization (WHO), 2020a. Cyanobacterial toxins: microcystins. Guidel. Drink. Qual. Guidel. Safe Recreat. Water Environ. 63.

Submitted by:   
 Mark T. Aubel, Ph.D.  
 Lab Director  
 Date: June 23, 2021

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**Cyano  
LAB**

## Potentially Toxicogenic (PTOX) Cyanobacteria Report

*Project: BlueGreen Water Tech*

Submitted to: Waleed Nasser  
 Organization: BlueGreen US Water Technologies, Inc  
 Address: #428, 1302 Waugh Dr., Houston, TX, 77019  
 Email: [waleed@bgtechs.com](mailto:waleed@bgtechs.com)  
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 Date Prepared: 14 June 2021\*  
 Prepared by: Alyssa Garvey

<u>Sample ID</u>	<u>Sites</u>	<u>Collected</u>
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34	C-43	6/6/21
35	C-43	6/6/21
36	C-43	6/6/21
37	C-43	6/6/21

*\*The samples were analyzed outside the holding time for unpreserved algal identification samples (<5 days), which may have impacted the results.*

### Method

A one mL aliquot of each non-preserved sample was prepared using a Sedgewick Rafter cell. The samples were scanned at 100X for the presence of potentially toxicogenic (PTOX) cyanobacteria using a Nikon Eclipse TE200 inverted microscope equipped with phase contrast optics. Higher magnification was used as necessary for identification and micrographs.

### Results

#### 31

The potentially toxicogenic (PTOX) cyanobacteria observed included *Dolichospermum* sp. (>40 filaments per mL; mostly partial), *Raphidiopsis raciborskii* (>30 filaments per mL), *Cuspidothrix* sp. (>20 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Microcystis* sp. (>10 colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**32**

The PTOX cyanobacteria observed included *Raphidiopsis raciborskii* (>25 filaments per mL), *Cuspidothrix* sp. (>15 filaments per mL), *Dolichospermum* sp. (>10 filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>5 filaments per mL), *Microcystis* sp. (>5 colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**33**

The sample was dominated by the PTOX cyanobacteria *Microcystis* spp. Other PTOX cyanobacteria observed included *Raphidiopsis raciborskii* (>10 filaments per mL), *Dolichospermum* sp. (>5 partial filaments per mL), *Cuspidothrix* sp. (>5 filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 2$  filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**34**

The PTOX cyanobacteria observed included *Dolichospermum* sp. (>15 filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (>10 filaments per mL), *Cuspidothrix* sp. (>5 filaments per mL), *Raphidiopsis raciborskii* ( $\geq 4$  filaments per mL), *Microcystis* sp. ( $\geq 4$  colonies per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**35**

The PTOX cyanobacteria observed included *Microcystis* sp. (>20 colonies per mL), *Cuspidothrix* sp. (>10 filaments per mL), *Raphidiopsis raciborskii* (>5 filaments per mL), *Dolichospermum* sp. (>5 filaments per mL; mostly partial), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 2$  filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**36**

The PTOX cyanobacteria observed included *Microcystis* sp. (>20 colonies per mL), *Raphidiopsis raciborskii* (>5 filaments per mL), *Cuspidothrix* sp. (>5 filaments per mL), *Dolichospermum* sp. ( $\geq 5$  partial filaments), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. (1 filament per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

**37**

The PTOX cyanobacteria observed included *Microcystis* sp. (>15 colonies per mL), *Raphidiopsis raciborskii* (>5 filaments per mL), *Dolichospermum* sp. ( $\geq 5$  partial filaments per mL), *Cuspidothrix* sp. ( $\geq 4$  filaments per mL), *Aphanizomenon/Chrysoosporum/Sphaerospermopsis* sp. ( $\geq 2$  filaments per mL), and *Pseudanabaena* sp. (<100 filaments per mL).

Potential toxin producing genera observed include:

Microcystins	Saxitoxins	Anatoxin-a	Cylindrospermopsin
<i>Pseudanabaena</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>	<i>Dolichospermum</i>
<i>Dolichospermum</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>	<i>Cuspidothrix</i>
<i>Microcystis</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>	<i>Aphanizomenon</i>
<i>Sphaerospermopsis</i>	<i>Chrysochlorum</i>	<i>Chrysochlorum</i>	<i>Chrysochlorum</i>
	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>	<i>Sphaerospermopsis</i>
	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>	<i>Raphidiopsis</i>

### Recommendations

Based on these observations and previous toxin data, analysis for microcystins and saxitoxins is recommended for **all** samples.

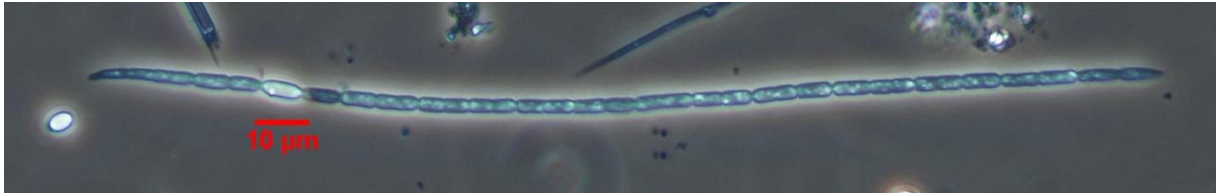
### Micrographs



*Dolichospermum* sp. at 400X (31)



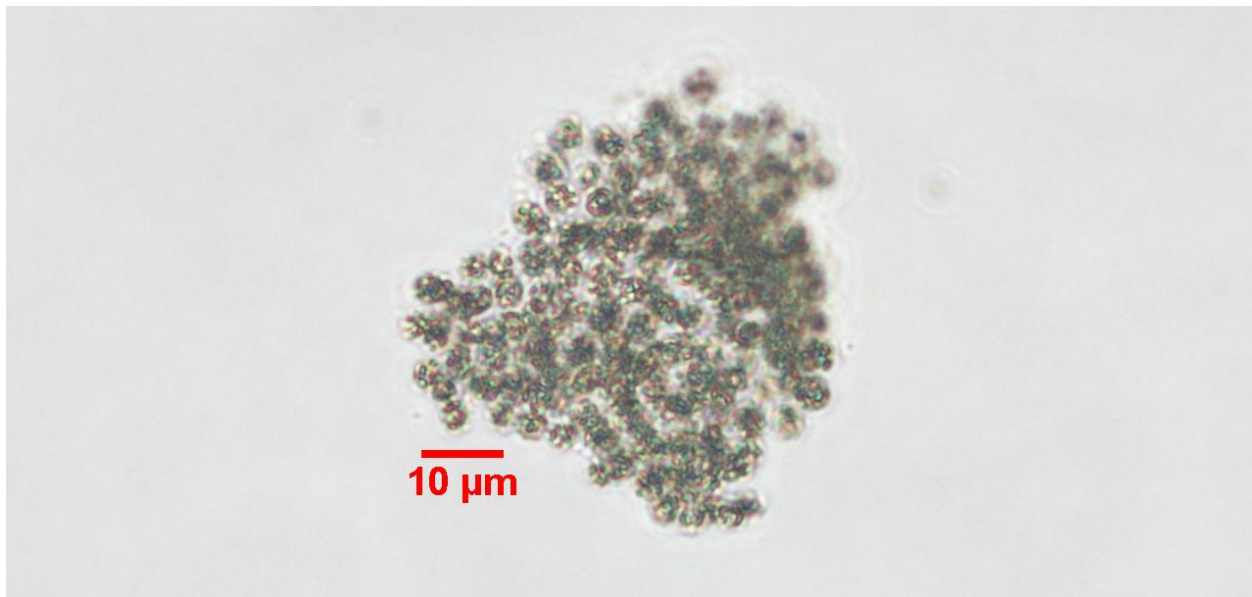
*Raphidiopsis raciborskii* at 400X (31)



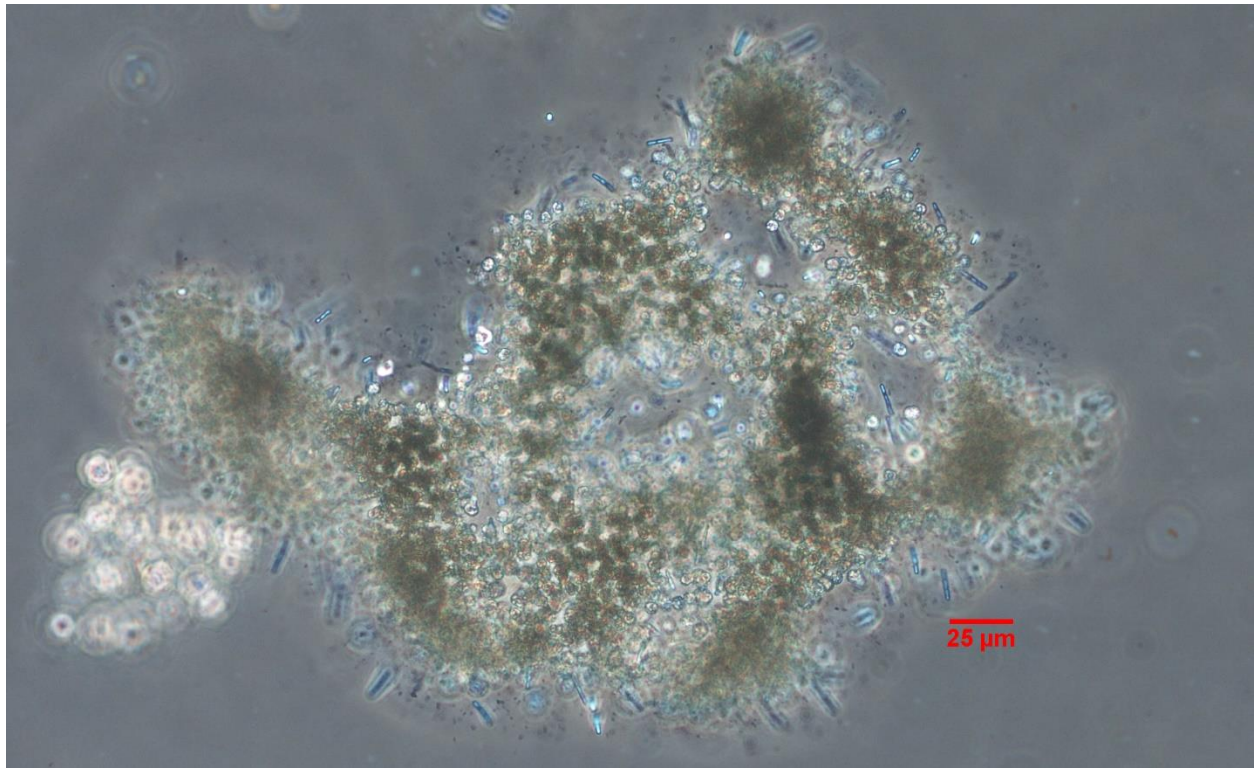
*Cuspidothrix* sp. at 400X (32)



*Aphanizomenon/Chrysochlorum/Sphaerospermopsis* sp. at 400X (32)



*Microcystis* sp. at 400X (33)



*Microcystis* sp. at 200X (33)



*Pseudanabaena* sp. at 400X (31)

Submitted by:

*Amanda Foss*

Amanda Foss, M.S.

Date:

June 14, 2021

*The results in this report relate only to the samples listed above.  
This report shall not be reproduced except in full without written approval of the laboratory.*

# Appendix D1



## CosmosID - Unlocking the Microbiome

Filename:	Sample_1_BWT2214
Size:	13.79 GiB
Reads:	68.593M
Date of Report:	2021-08-15 00:43:27

## Summary

Database Name	Hits	Status
Bacteria	172	Success
Antibiotic Resistance	1	Success
Fungi	72	Success
Protists	69	Success
Dark Matter (Beta)	163	Success
Phages	22	Success
Viruses	5	Success
Respiratory Virus	1	Success
Virulence Factors	6	Success



## Bacteria

Sample\_1\_BWT2214

## Fasta/q details

Total results

Metric	Value
Hit	0.920M

## Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis panniformis FACHB-1757	1638788	32328.68	49.02	69.08	80.14	94032
Microcystis aeruginosa PCC 9807	1160283	11157.42	16.92	26.42	53.35	28330
Microcystis sp. 0824	1502726	3230.66	4.90	12.04	24.75	11247
Microcystis viridis NIES-102	213615	2015.78	3.06	11.28	26.74	3458
Candidatus Fonsibacter ubiquis	1925548	1876.84	2.85	19.29	19.32	3066
Pseudomonas aeruginosa	287	1634.42	2.48	17.47	17.47	512
Phenylobacterium parvum	2201350	1576.03	2.39	11.42	11.42	27105
Sphingorhabdus contaminans	1343899	1468.72	2.23	11.92	12.00	22945
Clavibacter sp.	1871044	1439.88	2.18	13.14	13.14	4028
Dolichospermum circinale AWQC131C	398007	1102.30	1.67	8.61	11.74	5113
Synechococcus sp. CB0101	232348	753.29	1.14	5.78	5.89	10729
Limnohabitans sp. 103DPR2	1678129	665.97	1.01	3.38	5.12	3510
Pseudomonas sp. HAR-UPW-AIA-41	1985301	503.54	0.76	4.71	4.71	8010
Limnothrix sp. P13C2	1880902	483.26	0.73	3.01	4.91	54
Aestuariiirgva litoralis	2650924	448.98	0.68	3.56	3.56	12761
Pararheinheimeria texasensis DSM 17496	1123055	365.08	0.55	3.38	3.38	7283
unclassified Synechocystis	2640012	321.15	0.49	2.11	2.11	80
Flavobacterium fontis	1124188	243.88	0.37	2.55	2.59	3203
Fusobacterium nucleatum W1481	1408287	217.52	0.33	0.59	0.59	633
Neorickettsia	33993	211.05	0.32	6.06	6.06	13
Vulcanococcus limneticus LL	2025607	180.65	0.27	1.60	1.63	3793
Aquidulcibacter paucihalophilus	1978549	176.53	0.27	1.82	1.82	3306
Flectobacillus sp. BAB-3569	1509483	155.73	0.24	1.69	1.69	3096
Rhodobacter sp. CACIA14H1	1408890	155.29	0.24	1.16	1.35	4206
Novosphingobium ginsenosidimutans	1176536	153.78	0.23	1.59	1.59	2751
Cyanobium sp. NIES-981	1851505	144.84	0.22	1.09	1.39	2032
Staphylococcus aureus	1280	134.61	0.20	6.45	2.17	19
Candidatus Planktophila sp.	2175601	114.87	0.17	1.34	1.88	140
Polynucleobacter victoriensis	2049319	93.78	0.14	1.22	1.23	681
Methylocystis sp. ATCC 49242	622637	89.93	0.14	0.97	1.01	2674
Cyanobium usitatum str. Tous	2116684	88.23	0.13	0.89	0.95	1403
Caedibacter taeniospiralis	28907	82.54	0.13	1.17	1.17	476
Tabrizicola sp. TH137	2067452	74.07	0.11	0.71	0.90	2457
Bacteroidetes bacterium SCGC AAA027-G08	938698	63.69	0.10	0.84	0.80	334
Raphidiopsis brookii D9	533247	62.70	0.10	0.88	1.12	480
beta proteobacterium SCGC AAA028-K02	938797	60.00	0.09	0.98	0.98	188
Flavobacterium sasangense DSM 21067	1121896	57.50	0.09	0.80	0.93	584
Bacillus cereus group	86661	51.82	0.08	1.00	1.00	19
Inhella crocodyli	2499851	51.71	0.08	0.62	0.62	1444
actinobacterium SCGC AAA027-J17	932040	50.33	0.08	0.83	0.83	157
Acidovorax kalamii	2004485	49.29	0.07	0.62	0.76	1109
Porphyrobacter sp. LM 6	1896196	48.78	0.07	0.60	0.60	939
Methylocystis	133	47.51	0.07	1.28	3.96	10
Pseudanabaena sp. ABRG5-3	685565	46.79	0.07	0.57	0.58	854
Flavobacterium indicum GPTSA100-9 = DSM 17447	1094466	43.90	0.07	0.68	0.68	352
Cyanobium gracile PCC 6307	292564	41.22	0.06	0.47	1.01	845
Comamonadaceae	80864	40.01	0.06	0.69	0.69	11

Elstera cyanobacteriorum	2022747	38.16	0.06	0.48	0.48	1009
Limnohabitans curvus	323423	38.26	0.06	0.46	1.22	455
Aphanothece cf. minutissima CICALA 015	2107695	36.01	0.06	0.44	0.99	691
beta proteobacterium SCGC AAA027-I06	938781	35.59	0.05	0.44	0.44	418
Aphanizomenon flos-aquae 2012/KM1/D3	1532906	31.24	0.05	0.51	0.76	211
Lactiplantibacillus plantarum	1590	29.46	0.04	0.20	0.20	62
Candidatus Nanopelagicus limnes	1884634	28.10	0.04	0.51	0.83	119
Methylibium	316612	28.10	0.04	0.33	0.33	54
Silanimonas lenta DSM 16282	1123253	27.46	0.04	0.38	0.38	510
Flavobacterium sp. WWJ-16	2506421	26.73	0.04	0.38	0.45	468
Ideonella sp. KYPY4	1862385	26.27	0.04	0.31	0.31	1003
Rhizobium sp. MSSRF QS100	1522278	24.90	0.04	0.37	0.68	379
Cylindrospermopsis sp. CR12	1747196	24.31	0.04	0.42	0.79	131
Anabaena sp. WA102	1647413	24.53	0.04	0.41	0.66	195
Mycobacterium sp. M26	1762962	24.61	0.04	0.27	0.27	1114
Polynucleobacter cosmopolitanus	351345	20.87	0.03	0.37	0.38	172
Rubrivivax albus	2499835	21.01	0.03	0.26	0.28	845
Dolichospermum sp. UHCC 0315A	1914871	20.14	0.03	0.33	0.51	184
Aeromonas	642	20.46	0.03	0.54	0.58	13
Verrucomicrobia bacterium SCGC AAA027-I19	939126	20.32	0.03	0.25	0.25	501
actinobacterium SCGC AAA028-N15	938467	19.72	0.03	0.38	0.38	81
Pseudopuniceibacterium sediminis	2211117	19.67	0.03	0.65	0.65	772
Cylindrospermopsis raciborskii CS-505	533240	19.26	0.03	0.37	0.79	96
Limnohabitans parvus II-B4	1293052	18.45	0.03	0.24	0.27	428
Clostridiales	186802	17.57	0.03	1.68	1.68	2
Methylocystaceae	31993	18.12	0.03	0.32	0.32	22
Pseudomonas alcaligenes NBRC 14159	1215092	18.11	0.03	0.27	0.61	314
Rickettsiae	33988	17.41	0.03	100.00	100.00	34
Vogesella urethralis	2592656	16.98	0.03	0.24	0.25	524
Rhizobiales bacterium CCH10-E5	1768797	16.32	0.03	0.24	0.48	393
Dechloromonas sp. H13	2570193	16.19	0.03	0.21	0.22	542
Methylocystis parvus OBBP	1134912	15.60	0.02	0.20	0.24	638
Piscinibacter defluvii	1796922	15.80	0.02	0.21	0.27	635
Opiritaceae bacterium TAV3	278958	16.03	0.02	0.27	0.30	41
Hydrogenophaga sp. H7	1882399	15.09	0.02	0.20	0.20	574
Rubrivivax gelatinosus	28068	15.04	0.02	0.13	0.26	266
Crenobacter sp. GY 70310	2563443	15.35	0.02	0.22	0.22	449
alpha proteobacterium SCGC AAA027-C06	938624	14.34	0.02	0.38	0.47	28
Erythrobacter neustonensis	1112	14.35	0.02	0.23	0.23	290
Gemmobacter aquaticus	490185	13.71	0.02	0.19	0.19	445
Acidovorax sp. GW101-3H11	1813946	13.65	0.02	0.20	0.37	394
Aquabacterium pictum	2315236	14.17	0.02	0.18	0.19	596
Plesiomonas shigelloides	703	13.97	0.02	0.18	0.42	33
Clostridium	1485	13.34	0.02	3.41	5.45	6
Limnohabitans planktonicus II-D5	1293045	13.03	0.02	0.16	0.17	507
Curvibacter sp. PAE-UM	1714344	12.89	0.02	0.17	0.17	457
Methylomonas sp. MK1	1131552	12.98	0.02	0.29	0.28	631
Aquirufa antheringensis	2516559	13.17	0.02	0.21	0.21	241
Mycobacteriaceae	1762	12.72	0.02	0.32	0.32	10
alpha proteobacterium SCGC AAA487-M09	938672	12.24	0.02	0.41	0.34	17
Macromonas sp. BK-30	1843082	12.43	0.02	0.18	0.18	355
Piscinibacter aquaticus	392597	12.76	0.02	0.17	0.18	545
actinobacterium SCGC AAA027-L06	913338	12.04	0.02	0.26	0.33	39
Zhizhongheella caldifontis	1452508	11.89	0.02	0.17	0.21	406
Malikia granosa	263067	11.74	0.02	0.16	0.57	363

Novosphingobium kunmingense	1211806	11.07	0.02	0.15	0.15	417
Ramlibacter tataouinensis TTB310	365046	11.19	0.02	0.15	0.16	451
Inhella inkyongensis	392593	11.48	0.02	0.17	0.17	335
Ideonella sakaiensis	1547922	11.42	0.02	0.16	0.20	463
Burkholderiales bacterium JOSHI_001	864051	11.25	0.02	0.16	0.16	462
Opiritaceae bacterium TAV4	278959	10.99	0.02	0.19	0.23	38
Quisquiliibacterium sp. CC-CFT501	2498847	10.59	0.02	0.14	0.14	467
Rivibacter subsaxonicus	457575	10.47	0.02	0.14	0.14	435
Burkholderiales genera incertae sedis	224471	10.81	0.02	0.22	0.22	23
Paucibacter aquatile	2070761	10.58	0.02	0.14	0.14	444
actinobacterium SCGC AAA278-O22	932044	10.18	0.01	0.16	0.16	124
Gemmobacter sp. HYN0069	2169400	10.00	0.01	0.13	0.13	408
Holospira obtusa F1	1399147	10.17	0.01	0.19	0.19	96
Anaeromyxobacter	161492	9.76	0.01	0.16	0.16	69
Actinobacteria bacterium IMCC26077	1848755	9.39	0.01	0.14	0.14	164
Rubrivivax benzoatilyticus JA2 = ATCC BAA-35	987059	9.32	0.01	0.13	0.25	405
Cetobacterium somerae ATCC BAA-474	1319815	9.39	0.01	0.20	0.21	56
Gemmatimonas	173479	9.24	0.01	9.59	9.47	58
Runella sp. SP2	2268026	9.32	0.01	0.14	0.19	365
actinobacterium SCGC AAA028-A23	932036	8.63	0.01	0.34	0.37	11
actinobacterium acIB-AMD-7	1504322	8.53	0.01	0.23	0.38	31
Rhodobacter flagellatus	2593021	7.66	0.01	0.11	0.13	279
Burkholderia mallei	13373	8.14	0.01	0.24	0.16	15
Sulfurisoma sedimicola	1381557	7.84	0.01	0.12	0.12	263
Candidatus Accumulibacter sp. SK-12	1454001	7.67	0.01	0.11	0.12	336
Pseudanabaena biceps PCC 7429	927668	7.28	0.01	0.12	0.13	170
Clostridium perfringens	1502	7.57	0.01	0.26	0.26	9
alpha proteobacterium SCGC AAA028-C07	938639	6.99	0.01	0.20	0.23	22
Alphaproteobacteria bacterium SYSU XM001	2560861	7.09	0.01	0.10	0.10	317
Novosphingobium sp. B 225	1961849	6.94	0.01	0.11	0.11	225
Tibeticola sediminis	1917811	7.03	0.01	0.11	0.11	229
Ramlibacter sp. Leaf400	1736365	7.58	0.01	0.11	0.13	351
Ramlibacter rhizophilus	1781167	7.19	0.01	0.10	0.10	339
Simplicispira metamorpha	80881	7.17	0.01	0.11	0.11	252
Sphaerotilus natans subsp. natans DSM 6575	1286631	7.19	0.01	0.10	0.11	319
Aquincola tertiaricarbonis	391953	7.30	0.01	0.11	0.11	329
Flavohumibacter sp. SB-02	2676868	7.36	0.01	0.13	0.13	165
Exiguobacterium	33986	6.33	0.01	0.13	0.13	4
Schlegelella thermodepolymerans	215580	6.88	0.01	0.11	0.11	269
Tepidicella xavieri	360241	6.62	0.01	0.10	0.11	200
Acinetobacter sp. VT 511	1675902	6.67	0.01	0.19	0.56	25
Ignavibacteriales	795748	6.36	0.01	16.67	16.67	13
Flavobacteria bacterium BAL38	391598	6.62	0.01	0.17	0.23	41
Acidovorax defluvii	86669	6.26	<0.01	0.11	0.12	130
Legionella donaldsonii	45060	5.92	<0.01	0.11	0.10	101
Aeromonas salmonicida subsp. achromogenes AS03	1233098	6.09	<0.01	0.21	0.21	26
actinobacterium SCGC AAA278-I18	938557	5.34	<0.01	0.15	0.15	24
Holospira curviuscula	1082868	5.04	<0.01	0.12	0.12	47
Bacillus sp. JEM-1	1977090	4.32	<0.01	0.32	0.32	4
alpha proteobacterium SCGC AAA027-J10	938631	4.84	<0.01	0.24	0.28	9
alpha proteobacterium SCGC AAA028-D10	938641	4.52	<0.01	0.14	0.19	17
Methylophilus	16	4.43	<0.01	5.26	5.26	2
unclassified Candidatus Accumulibacter	2619054	4.57	<0.01	0.10	0.10	2
Xanthomonadaceae bacterium JGI 0001002-F18	1094969	4.87	<0.01	0.38	0.38	19
Mycobacterium	1763	3.89	<0.01	0.10	0.10	18

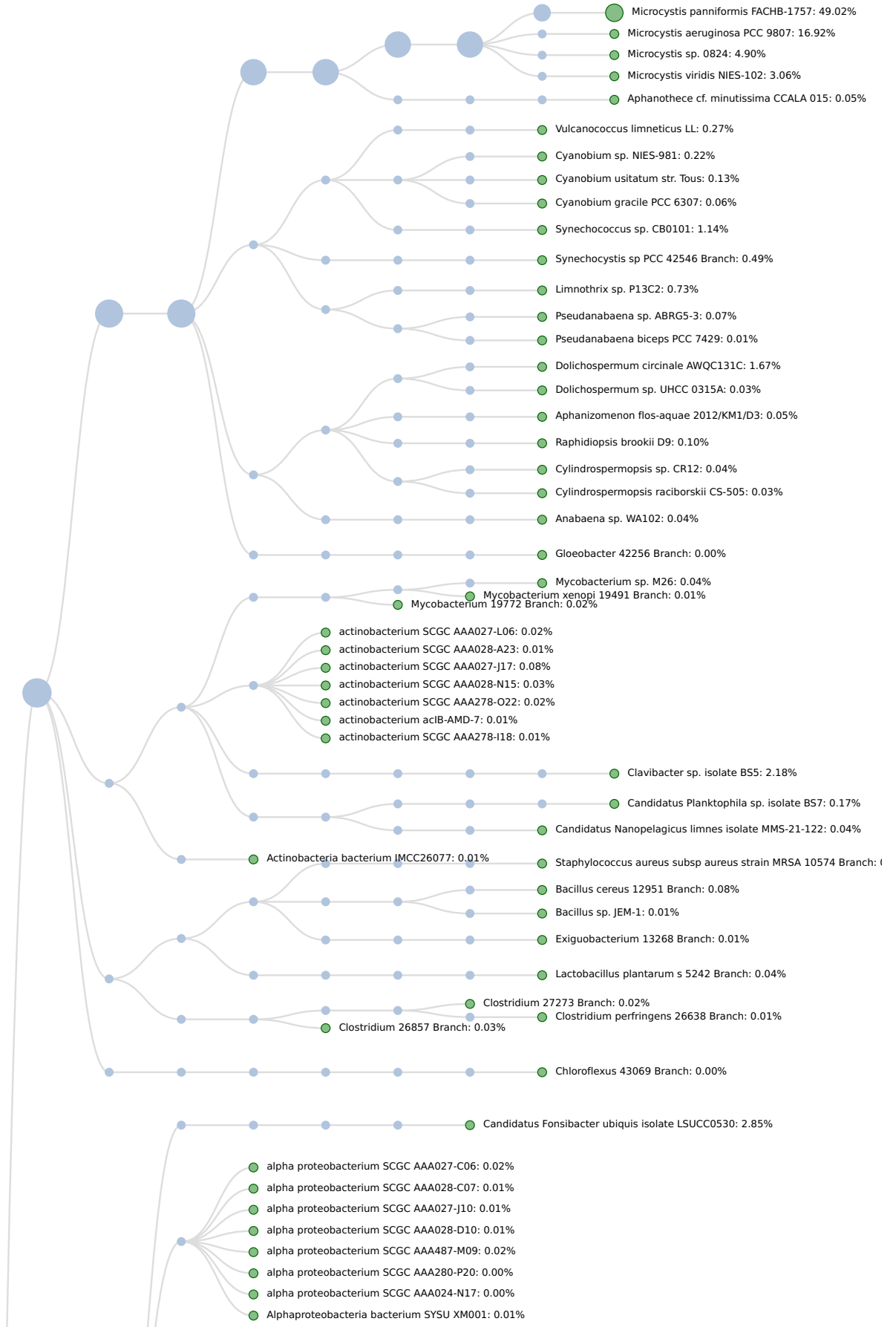
Comamonadaceae bacterium JGI 0001013-A16	1286843	3.83	<0.01	0.11	0.11	22
Acinetobacter schindleri	108981	3.96	<0.01	0.14	0.20	15
Wolbachia endosymbiont of Drosophila ananassae	307502	3.32	<0.01	0.26	0.20	13
Comamonas	283	3.49	<0.01	0.12	0.13	9
Burkholderiales	80840	3.32	<0.01	11.11	11.11	7
Ferrovum sp. JA12	1356299	3.27	<0.01	0.11	0.11	82
Chitinilyticum	551208	3.27	<0.01	0.12	0.12	5
Escherichia coli MS 110-3	749536	3.52	<0.01	0.63	0.47	3
Alphaproteobacteria	28211	2.31	<0.01	0.10	0.10	8
Gloeobacter	33071	2.18	<0.01	2.01	2.01	11
Chloroflexus	1107	1.87	<0.01	6.90	6.90	4
alpha proteobacterium SCGC AAA280-P20	938665	1.88	<0.01	0.12	0.12	5
unclassified Verrucomicrobia	417295	1.91	<0.01	0.14	0.14	2
alpha proteobacterium SCGC AAA024-N17	938623	1.01	<0.01	0.18	0.22	9
unclassified Opitutaceae	278955	1.28	<0.01	1.27	1.66	7

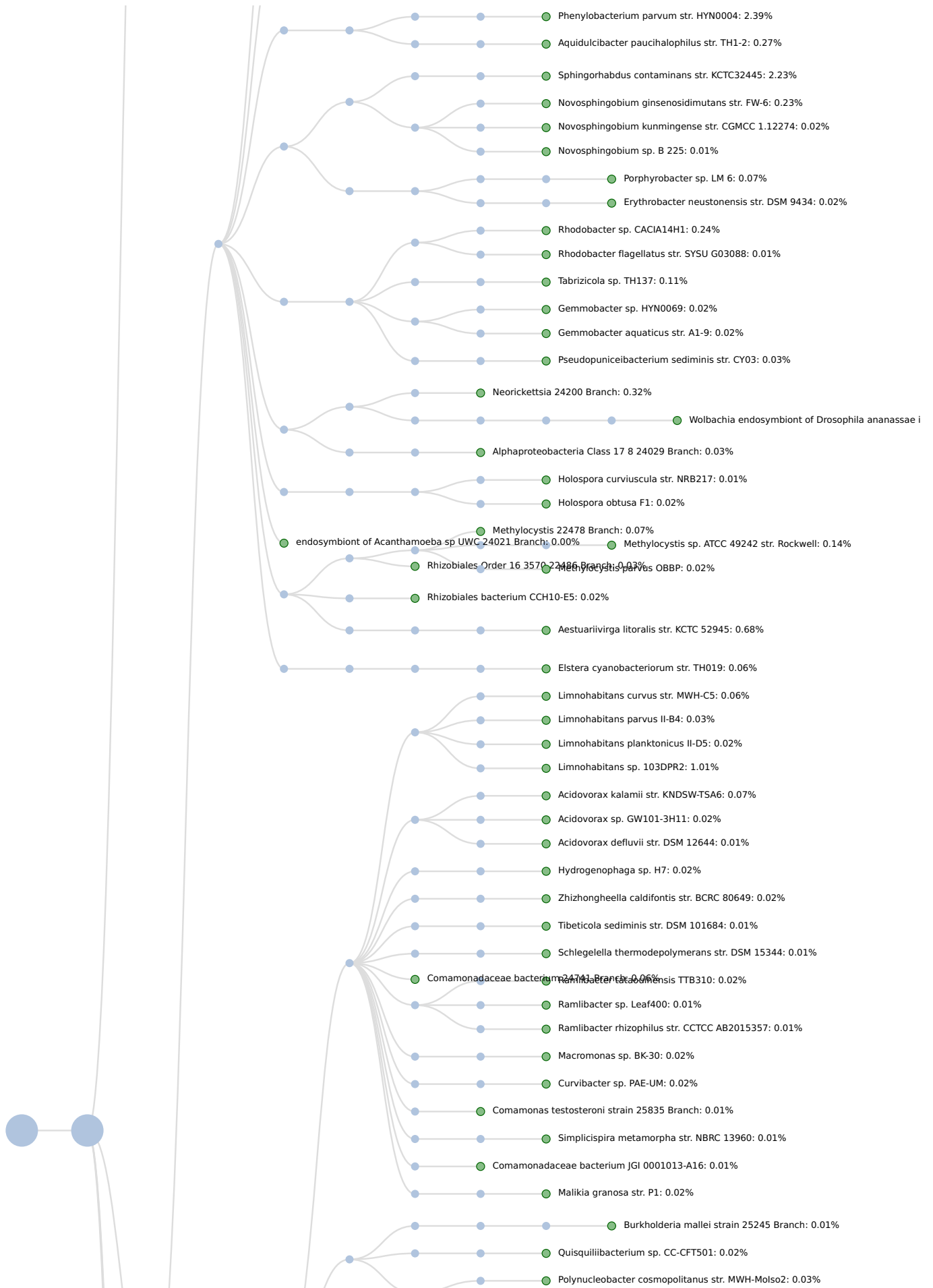
Bacteria

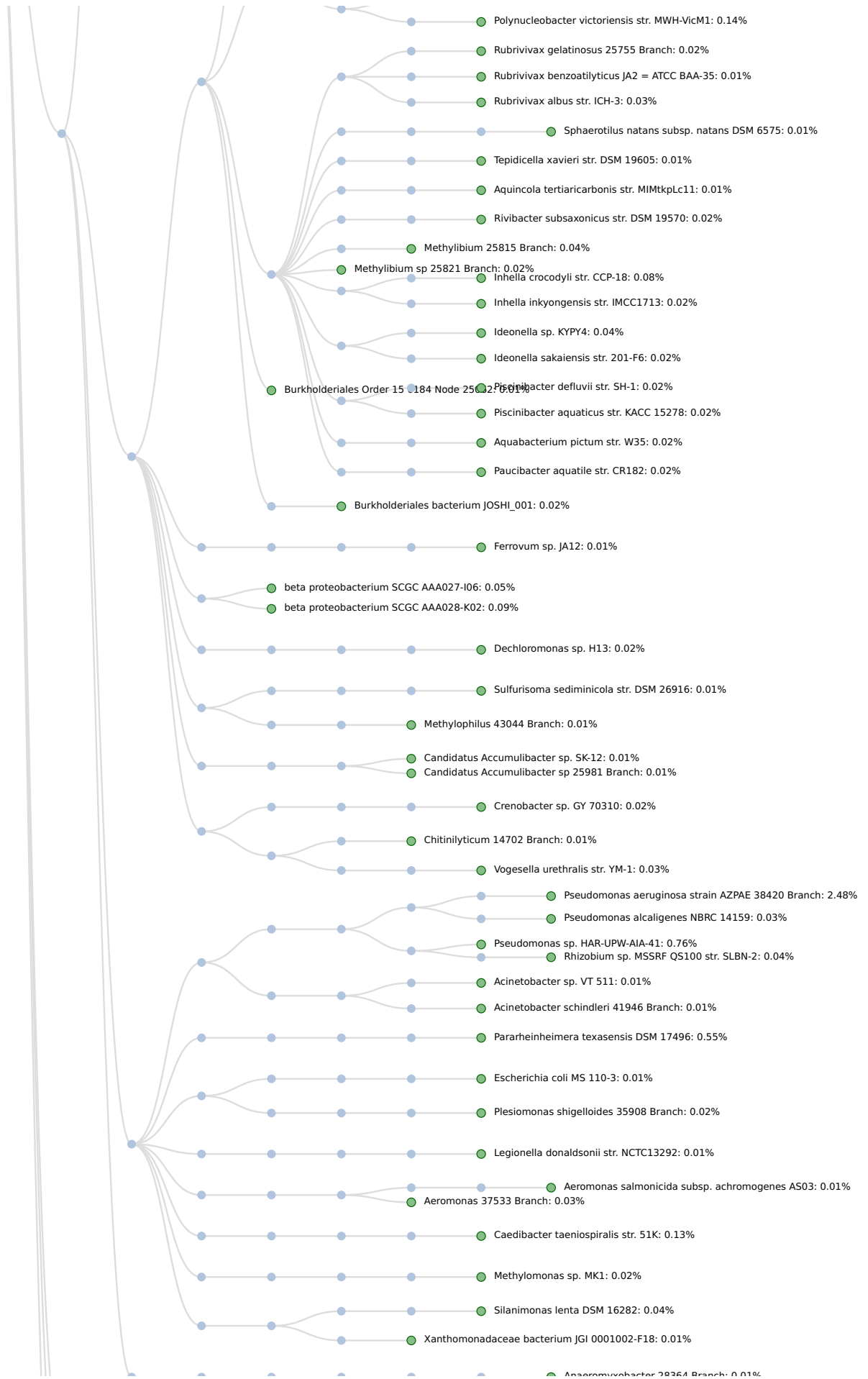
Tree Chart Taxonomy

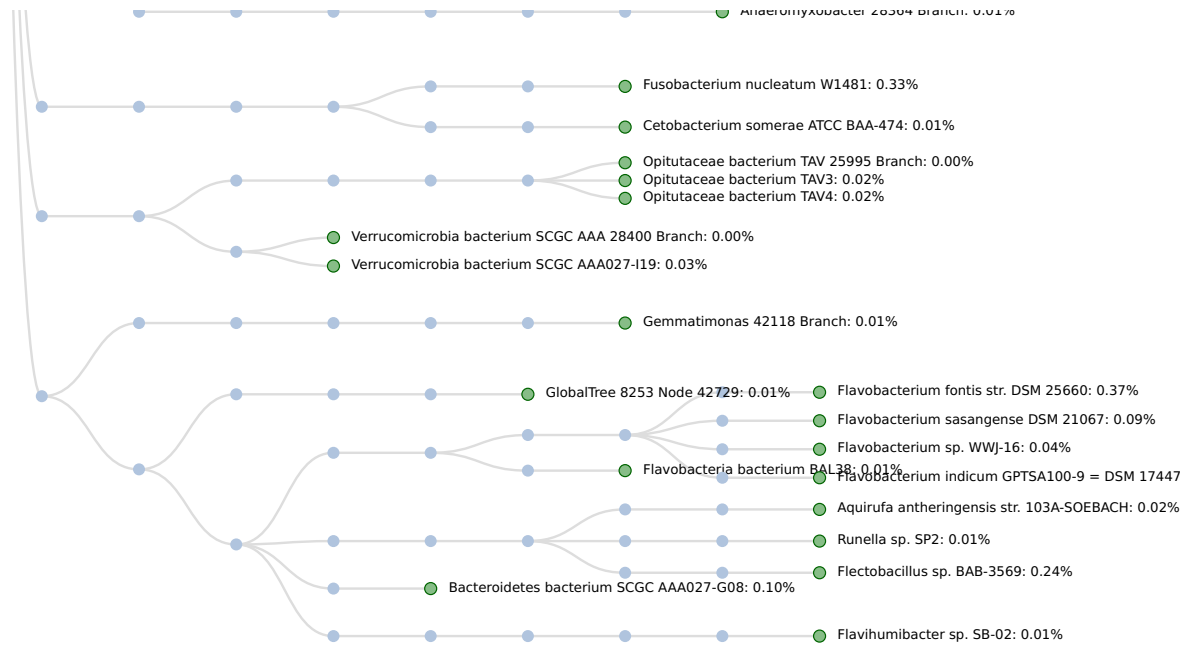
Sample\_1\_BWT2214

Total results









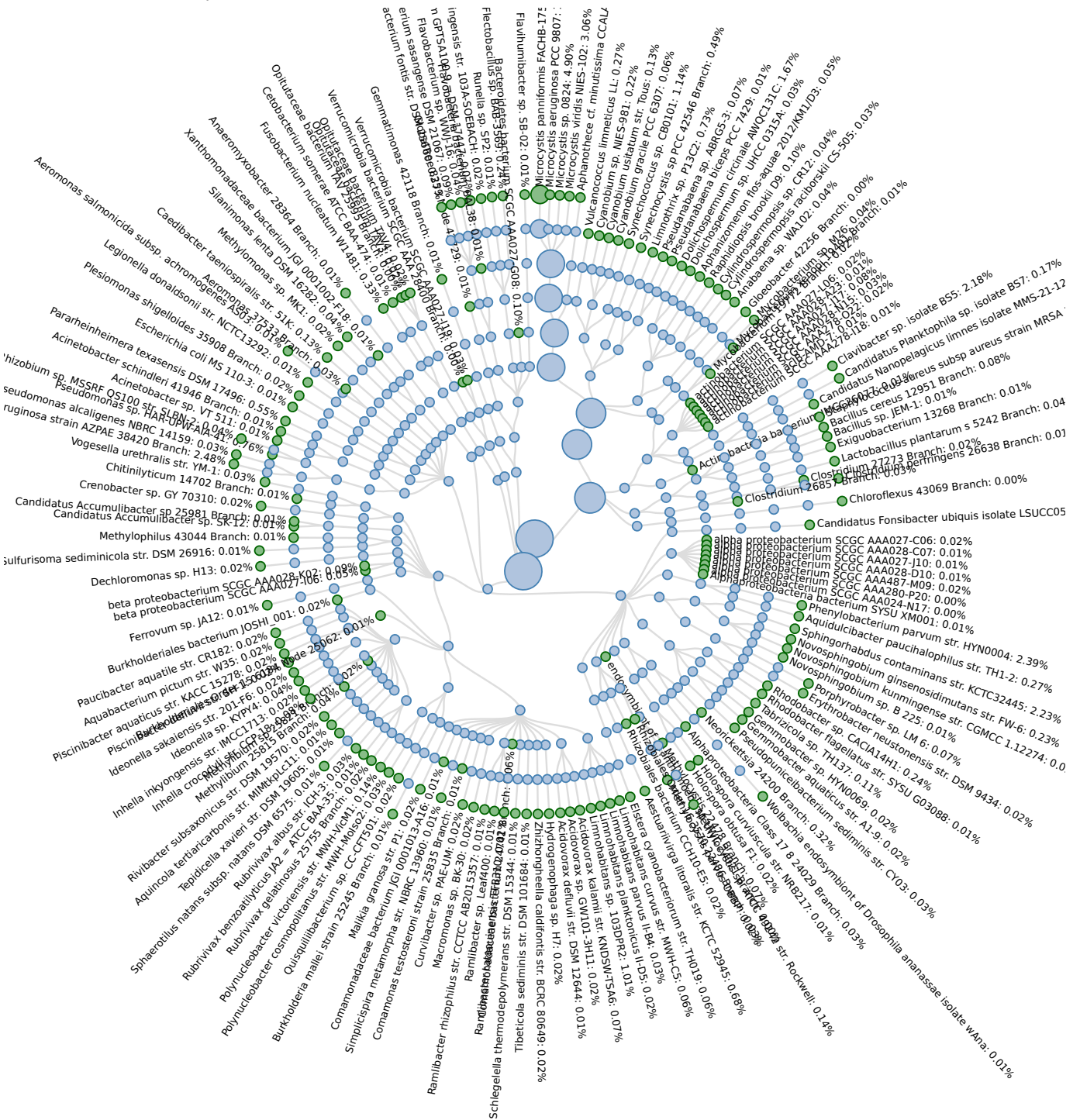


Bacteria

Radial Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results



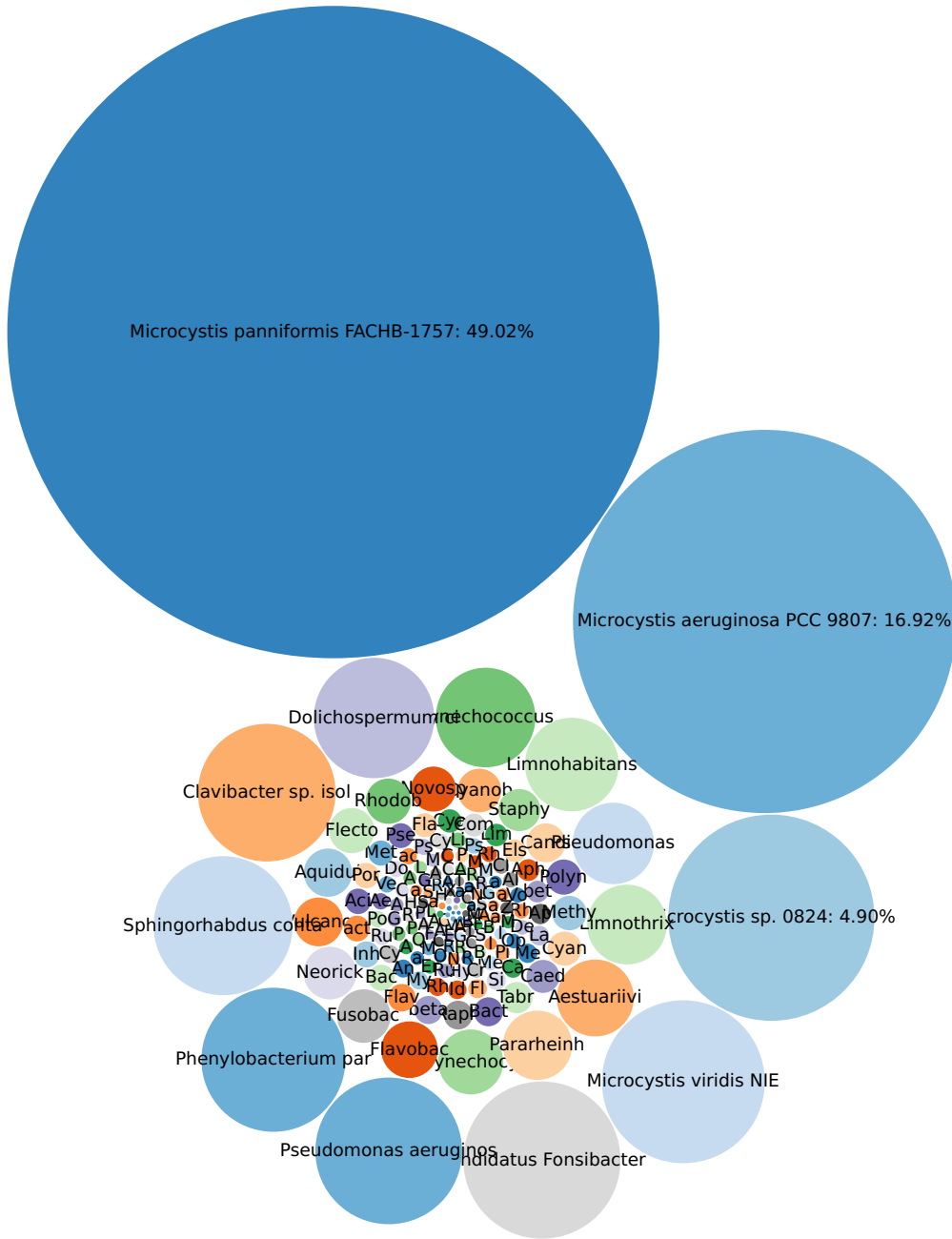


Bacteria

Bubble Chart Taxonomy

Sample\_1\_BWT2214

Total results



## Antibiotic Resistance

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	115

Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Aminoglycoside GENE aadA1 1 X02340	-	1048.98	100.00	35.94	13.53	130

## Fungi

Sample\_1\_BWT2214

## Fasta/q details

Total results

Metric	Value
Hit	58.204k

## Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Onygenales	33183	39.71	23.49	0.34	0.54	1508
Clavaria fumosa	264083	8.27	4.89	0.10	0.30	452
Fusarium subglutinans	42677	6.92	4.09	0.08	0.05	412
Brettanomyces bruxellensis	5007	5.35	3.16	0.07	0.08	325
Puccinia arachidis	333523	5.26	3.11	0.08	0.13	275
Trichosporon inkin	82517	4.35	2.57	0.02	0.02	454
Cantharellus cibarius	36066	4.33	2.56	0.06	0.08	257
Metschnikowia cerradonensis	390697	4.28	2.53	0.02	0.02	463
Morchella eximia	1582338	3.91	2.31	0.03	0.02	368
Cantharellus lutescens	104198	3.33	1.97	0.03	0.24	277
Epichloe sylvatica	79593	3.11	1.84	0.04	0.03	234
Phycomyces blakesleeanus NRRL 1555(-)	763407	3.02	1.79	0.03	0.03	277
Ceraceosorus bombacis	401625	2.80	1.66	0.02	0.01	287
Escovopsis weberi	150374	2.76	1.63	0.03	0.01	258
Ceratocystis smalleyi	312343	2.69	1.59	0.02	0.01	290
Epichloe bromicola	79588	2.62	1.55	0.02	0.01	278
Ophiocordyceps polyrhachis-furcata BCC 54312	1330021	2.45	1.45	0.02	0.02	240
Quambalaria eucalypti	363177	2.25	1.33	0.03	0.03	197
Ophiocordyceps unilateralis	268505	2.15	1.27	0.01	0.01	258
Chaetothyriales sp. CBS 132003	2249419	2.14	1.27	0.03	0.03	180
Ophiocordyceps camponoti-floridani	2030778	2.11	1.25	0.03	0.02	185
Escovopsis sp. AC	2027528	1.95	1.15	0.03	0.02	136
Geosmithia morbida	1094350	1.92	1.14	0.02	0.02	201
Ceratocystiopsis brevicomis	553390	1.89	1.12	0.03	0.03	149
Metschnikowia santaceciliae	197674	1.88	1.11	0.02	0.01	201
Blastomyces emzantsi	2723674	1.86	1.10	0.03	0.04	118
Sphaerulina populicola P02.02b	1136490	1.80	1.06	0.02	0.01	175
Melampsora pinitorqua Mpini7	1298852	1.78	1.05	0.02	0.03	155
Caulochytrium protostelioides	1555241	1.75	1.03	0.03	0.03	139
Metschnikowia continentalis	73517	1.72	1.02	0.01	0.01	192
Epichloe amarillans E57	1037526	1.69	1.00	0.02	0.02	148
Herpomyces periplanetae	1749284	1.62	0.96	0.01	0.01	192
Claviceps paspali	40601	1.48	0.88	0.01	0.02	162
Lentinus polychrous	292559	1.35	0.80	0.04	0.04	171
Cokeromyces recurvatus B5483	1357676	1.35	0.80	0.01	0.01	152
Ceraceosorus guamensis	1522189	1.33	0.79	0.02	0.01	846
Epichloe festucae FI1	877507	1.26	0.74	0.01	0.01	149
Epichloe uncinata	5050	1.25	0.74	0.02	0.01	471
Tuber microsphaerosporum	1455713	1.25	0.74	0.02	0.03	103
Apiotrichum gamsii	1105092	1.22	0.72	0.01	0.01	140
Mortierella sp. BCC40632	2613877	1.20	0.71	0.02	0.02	121
Lodderomyces elongisporus NRRL YB-4239	379508	1.17	0.69	0.02	0.02	88
Absidia glauca	4829	1.16	0.69	0.01	0.01	120
Metschnikowia sp. 00-154.1	1807683	1.13	0.67	0.02	0.02	99
Metschnikowia matae var. maris	1697387	1.04	0.61	0.02	0.01	99
Balansia obtecta B249	1405085	0.99	0.59	0.01	0.01	108
Epichloe elymi	55200	0.98	0.58	0.01	0.01	103

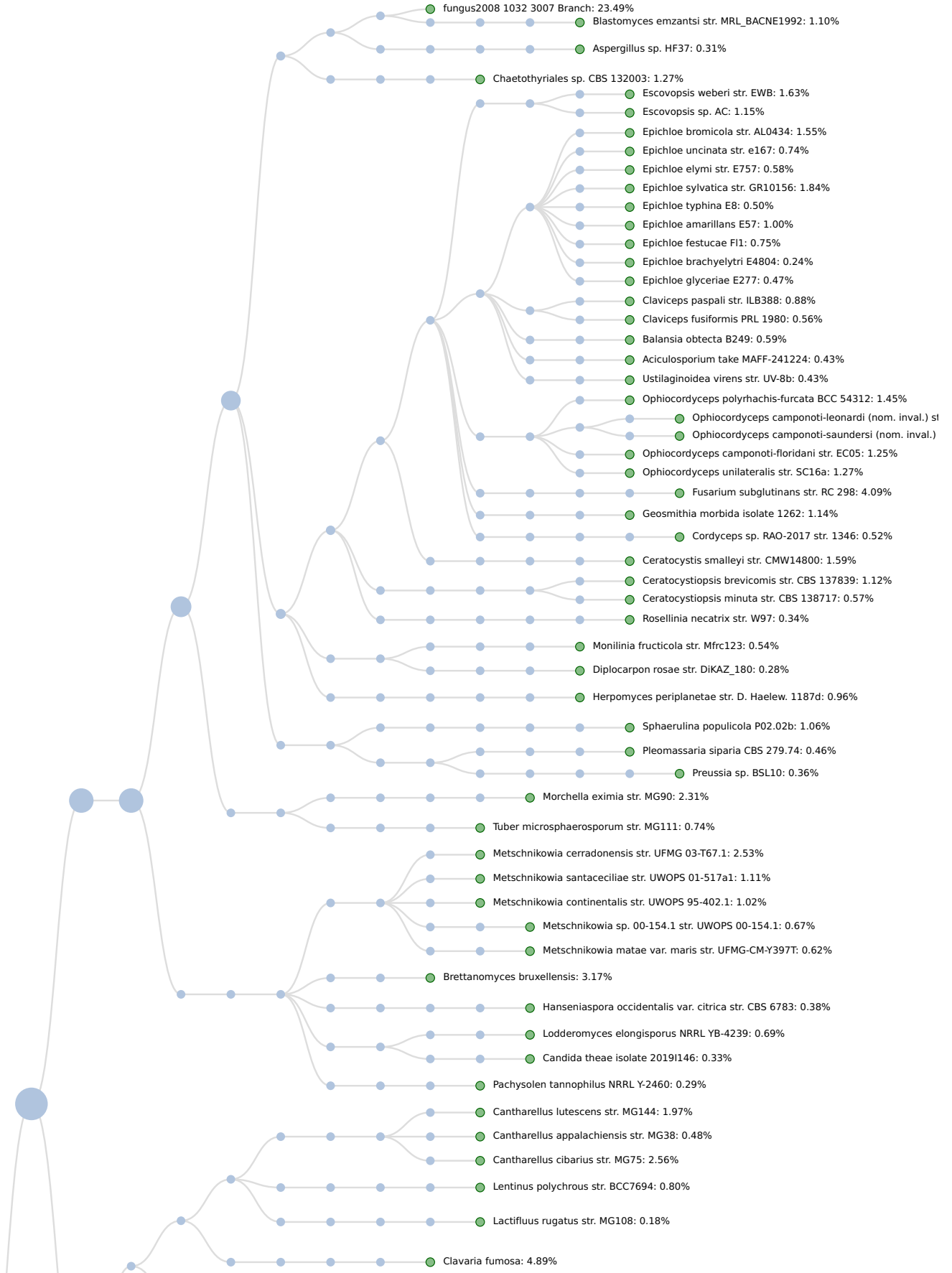
<i>Ceratocystiopsis minuta</i>	360147	0.97	0.57	0.01	0.02	102
<i>Claviceps fusiformis</i> PRL 1980	1036761	0.94	0.56	0.01	0.01	89
<i>Monilinia fructicola</i>	38448	0.92	0.54	0.02	0.01	83
<i>Cordyceps</i> sp. RAO-2017	2004951	0.88	0.52	0.02	0.02	78
<i>Anthracoystis flocculosa</i> PF-1	1277687	0.85	0.50	0.01	0.02	85
<i>Epichloe typhina</i> E8	1227655	0.84	0.50	0.01	0.01	83
<i>Cantharellus appalachiensis</i>	409893	0.81	0.48	0.01	0.19	71
<i>Epichloe glyceriae</i> E277	1035635	0.80	0.47	0.02	0.02	65
<i>Violaceomyces palustris</i>	1673888	0.80	0.47	0.01	0.01	77
<i>Pleomassaria siparia</i> CBS 279.74	1314801	0.78	0.46	0.01	0.01	75
<i>Microbotryum lychnidis-dioicae</i> p1A1 Lamole	683840	0.78	0.46	0.04	0.04	19
<i>Pecoramyces ruminatum</i>	1987568	0.76	0.45	0.01	0.01	76
<i>Aciculosporium take</i> MAFF-241224	1036760	0.72	0.43	0.01	0.02	62
<i>Ustilaginoidea virens</i>	1159556	0.72	0.43	0.01	0.01	70
<i>Hanseniaspora occidentalis</i> var. <i>citrica</i>	331638	0.65	0.39	0.01	0.01	273
<i>Preussia</i> sp. BSL10	1712568	0.60	0.36	0.02	0.02	36
<i>Rosellinia necatrix</i>	77044	0.57	0.34	0.01	0.01	56
<i>Candida theae</i>	1198502	0.56	0.33	0.01	0.01	46
<i>Ophiocordyceps camponoti-leonardi</i> (nom. inval.)	2039875	0.54	0.32	0.01	0.01	40
<i>Aspergillus</i> sp. HF37	1960876	0.53	0.31	0.01	0.01	139
<i>Ophiocordyceps camponoti-saundersi</i> (nom. inval.)	2039874	0.49	0.29	0.01	0.01	32
<i>Pachysolen tannophilus</i> NRRL Y-2460	669874	0.49	0.29	0.01	0.01	25
<i>Diplocarpon rosae</i>	946125	0.48	0.28	0.01	0.01	244
<i>Epichloe brachyelytri</i> E4804	1036762	0.41	0.24	0.01	0.01	88
<i>Lactifluus rugatus</i>	1837266	0.31	0.18	0.01	0.01	43

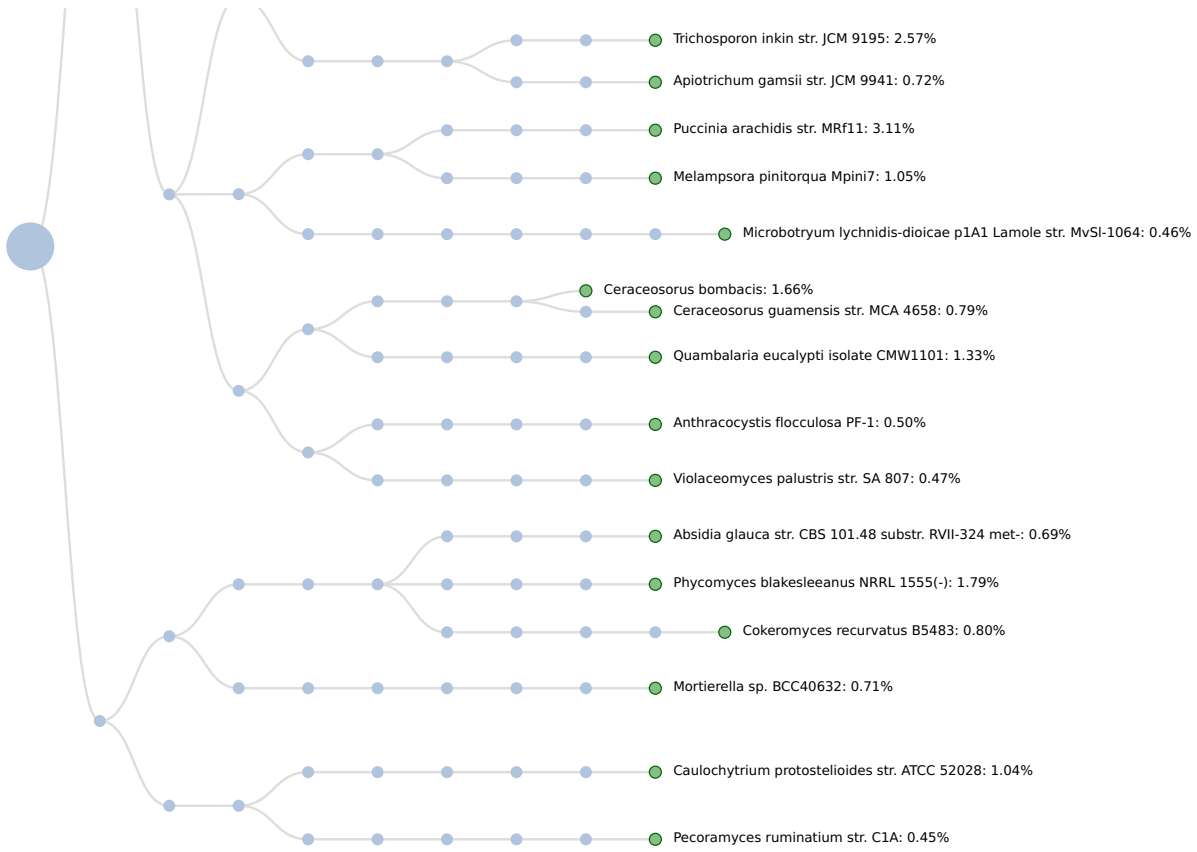
Fungi

Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results









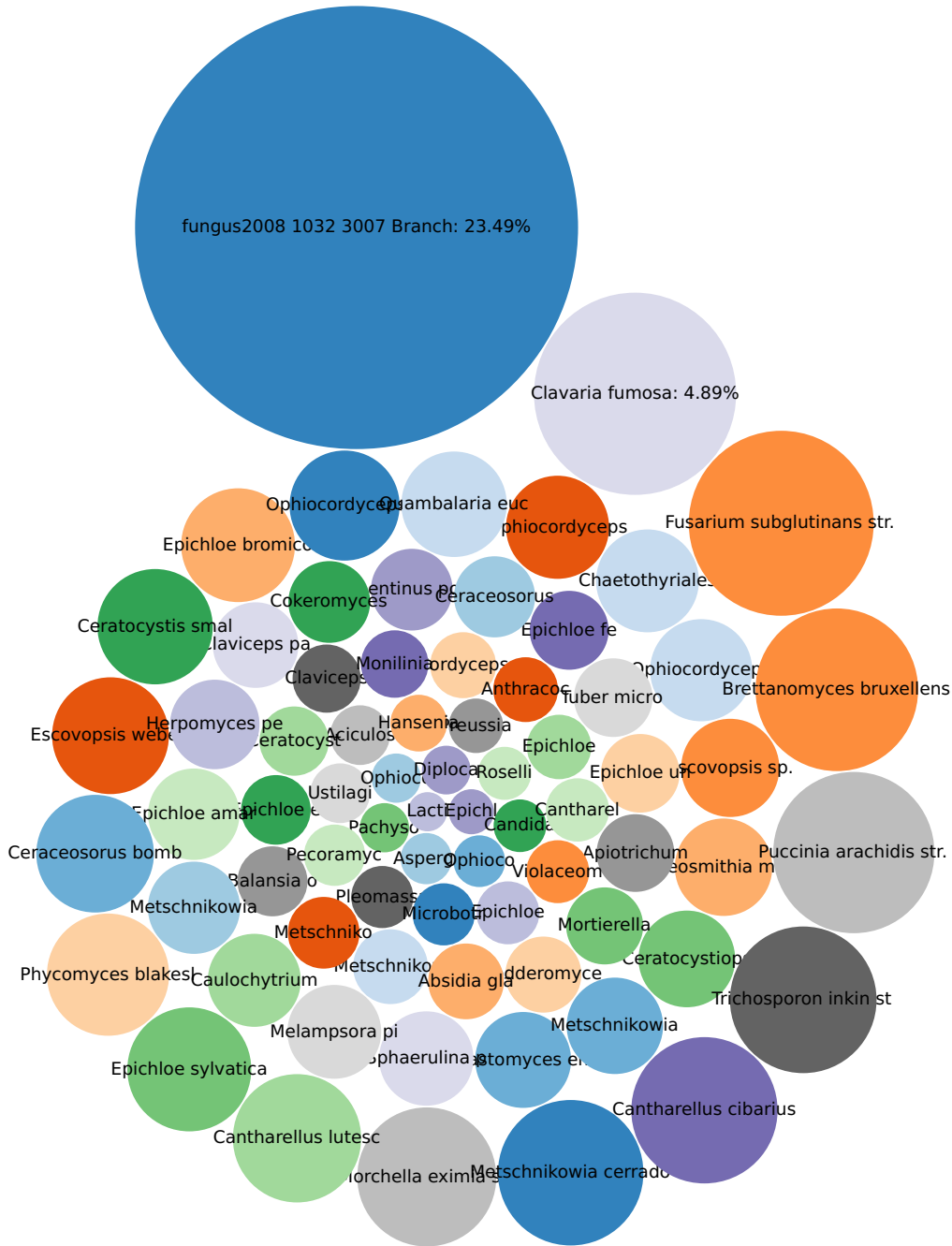


Fungi

Bubble Chart Taxonomy

Sample\_1\_BWT2214

Total results



## Protists

Sample\_1\_BWT2214

## Fasta/q details

Total results

Metric	Value
Hit	0.120M

## Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Neobalantidium coli	71585	5509.94	94.68	27.12	27.12	297
Salpingoeca rosetta	946362	42.00	0.72	0.33	2.17	2278
Acanthamoeba polyphaga	5757	21.82	0.38	0.20	0.18	1217
Fonticula alba	691883	16.69	0.29	0.10	0.24	1276
Symbiodinium minutum Mf 1.05b.01	1280413	14.95	0.26	0.09	1.29	1179
Acanthamoeba quina	211522	13.57	0.23	0.11	0.18	936
Pseudoperonospora cubensis	143453	12.49	0.21	0.15	0.21	605
Toxoplasma gondii p89	943119	10.04	0.17	0.02	0.03	411
Leishmania major strain LV39c5	860569	9.85	0.17	0.12	0.06	388
Bigelowiella natans CCMP2755	753081	8.72	0.15	0.08	0.53	621
Schizochytrium sp. CCTCC M209059	1573607	8.26	0.14	0.05	0.20	734
Thalassiosira pseudonana CCMP1335	296543	8.07	0.14	0.03	0.07	821
Reticulomyxa filosa	46433	8.05	0.14	0.06	0.16	631
Physarum polycephalum	5791	7.54	0.13	0.07	0.57	560
Guillardia theta CCMP2712	905079	7.37	0.13	0.05	0.29	653
Acanthamoeba pearcei	65662	6.74	0.12	0.06	0.17	193
Chromera velia	505693	6.76	0.12	0.07	0.94	475
Acanthamoeba lugdunensis	61605	6.67	0.11	0.07	0.20	458
Acanthamoeba mauritaniensis	196912	5.78	0.10	0.07	0.25	373
Acanthamoeba castellanii str. Neff	1257118	5.62	0.10	0.05	0.20	453
Hemiselms andersenii	464988	5.44	0.09	0.22	0.22	10
Acanthamoeba rhyodes	32599	3.99	0.07	0.05	0.22	279
Leishmania gerbilli	40285	3.78	0.07	0.04	0.05	283
Emiliana huxleyi CCMP1516	280463	3.79	0.07	0.04	0.48	296
Leishmania turanica	62297	3.66	0.06	0.05	0.05	229
Leishmania aethiopica L147	1206056	3.62	0.06	0.04	0.04	281
Acanthamoeba healyi	65661	3.50	0.06	0.03	0.33	297
Hammondia hammondi	99158	3.12	0.05	0.02	0.05	316
Nannochloropsis oceanica	145522	3.17	0.05	0.03	0.18	261
Sarcocystis neurona	42890	3.11	0.05	0.04	0.04	125
Leishmania infantum JPCM5	435258	3.05	0.05	0.04	0.03	207
Nannochloropsis gaditana CCMP526	1093141	2.94	0.05	0.04	0.04	177
Plasmodium gaboni	647221	2.59	0.04	0.01	0.03	150
Leishmania donovani	5661	2.42	0.04	0.03	0.02	182
Thecamonas trahens ATCC 50062	461836	2.45	0.04	0.02	0.08	257
Acanthamoeba palestiniensis	28015	2.24	0.04	0.03	0.28	161
Capsaspora owczarzaki ATCC 30864	595528	2.21	0.04	0.02	0.04	237
Cryptosporidium hominis	237895	1.83	0.03	0.02	0.01	109
Stylonychia lemnae 2x8/2	755200	1.78	0.03	0.04	0.05	51
Dictyostelium firmibasis	79012	1.71	0.03	0.02	0.03	123
Leishmania tropica L590	1206058	1.70	0.03	0.02	0.04	139
Leishmania arabica	40284	1.64	0.03	0.02	0.03	138
Entamoeba dispar SAW760	370354	1.55	0.03	0.03	0.03	74
Dictyostelium intermedium	361076	1.43	0.03	0.02	0.04	103
Paramecium biaurelia	65126	1.47	0.03	0.02	0.03	112
Cryptosporidium muris RN66	441375	1.37	0.02	0.02	0.02	64
Plasmodium reichenowi	5854	1.32	0.02	0.01	0.02	110

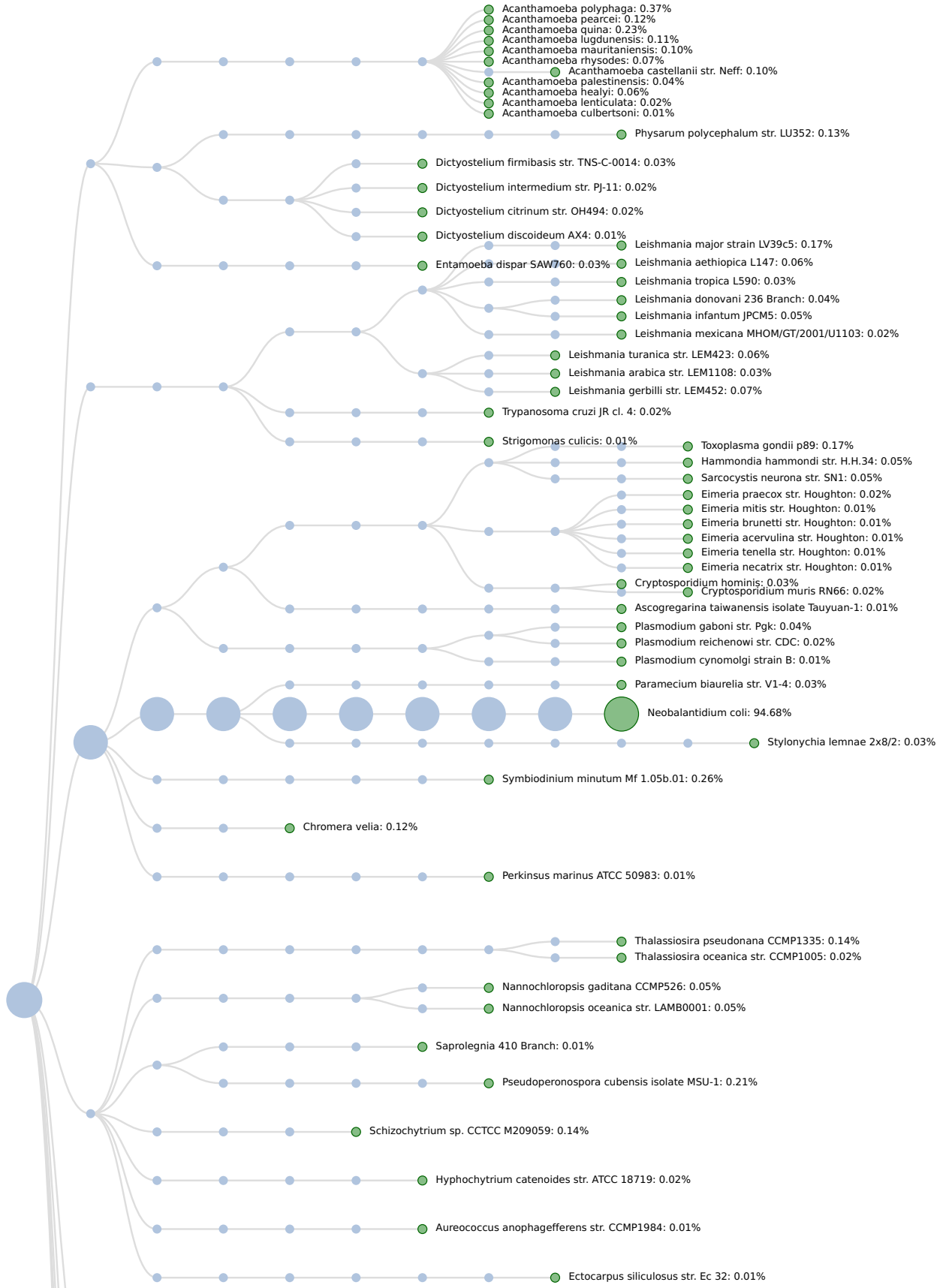
<i>Leishmania mexicana</i> MHOM/GT/2001/U1103	929439	1.19	0.02	0.01	0.01	129
<i>Hyphochytrium catenoides</i>	42384	1.19	0.02	0.01	0.03	131
<i>Dictyostelium citrinum</i>	361072	1.13	0.02	0.01	0.02	82
<i>Trypanosoma cruzi</i> JR cl. 4	914063	1.11	0.02	0.02	0.01	85
<i>Eimeria praecox</i>	51316	1.08	0.02	0.01	0.16	116
<i>Monosiga brevicollis</i> MX1	431895	1.13	0.02	0.02	0.07	163
<i>Acanthamoeba lenticulata</i>	29196	0.97	0.02	0.01	0.07	106
<i>Dictyostelium discoideum</i> AX4	352472	0.87	0.01	0.02	0.02	59
<i>Thalassiosira oceanica</i>	159749	0.89	0.01	0.01	0.06	629
<i>Saprolegnia</i>	4769	0.86	0.01	0.02	0.02	32
<i>Ascogregarina taiwanensis</i>	158379	0.74	0.01	0.01	0.02	69
<i>Strigomonas culicis</i>	28005	0.68	0.01	0.02	0.02	115
<i>Eimeria brunetti</i>	51314	0.72	0.01	0.01	0.21	66
<i>Aureococcus anophagefferens</i>	44056	0.71	0.01	0.01	0.08	68
<i>Ectocarpus siliculosus</i>	2880	0.70	0.01	0.01	0.26	55
<i>Plasmodium cynomolgi</i> strain B	1120755	0.65	0.01	0.01	0.02	53
<i>Perkinsus marinus</i> ATCC 50983	423536	0.64	0.01	0.01	0.06	66
<i>Eimeria acervulina</i>	5801	0.60	0.01	0.01	0.15	52
<i>Acanthamoeba culbertsoni</i>	43142	0.54	<0.01	0.01	0.07	43
<i>Eimeria necatrix</i>	51315	0.50	<0.01	0.01	0.15	34
<i>Eimeria mitis</i>	44415	0.47	<0.01	0.01	0.21	32
<i>Eimeria tenella</i>	5802	0.44	<0.01	0.01	0.10	32

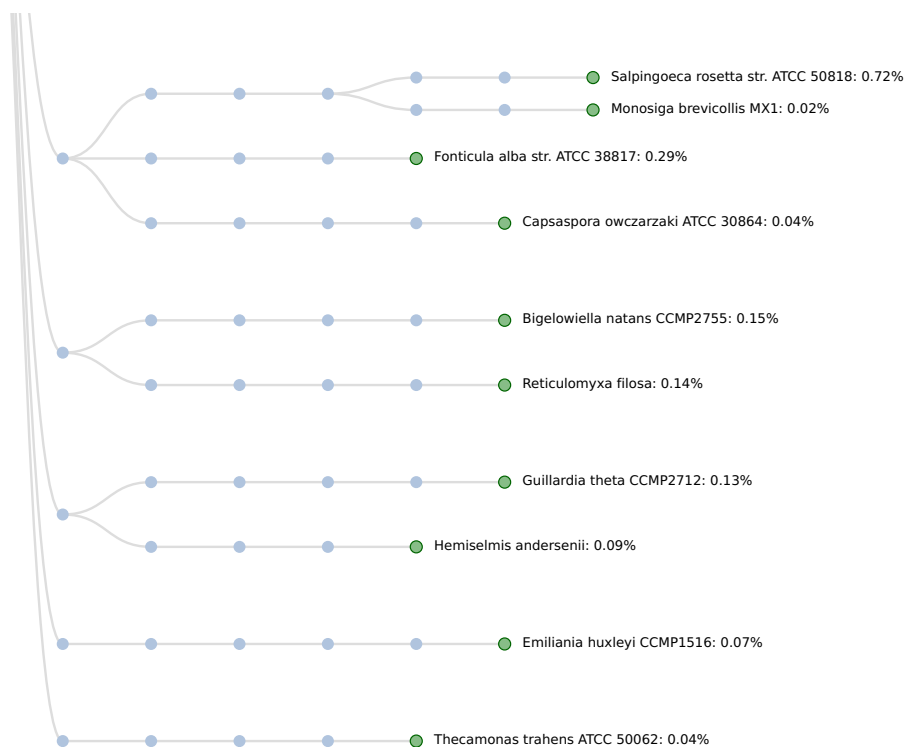
Protists

Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results



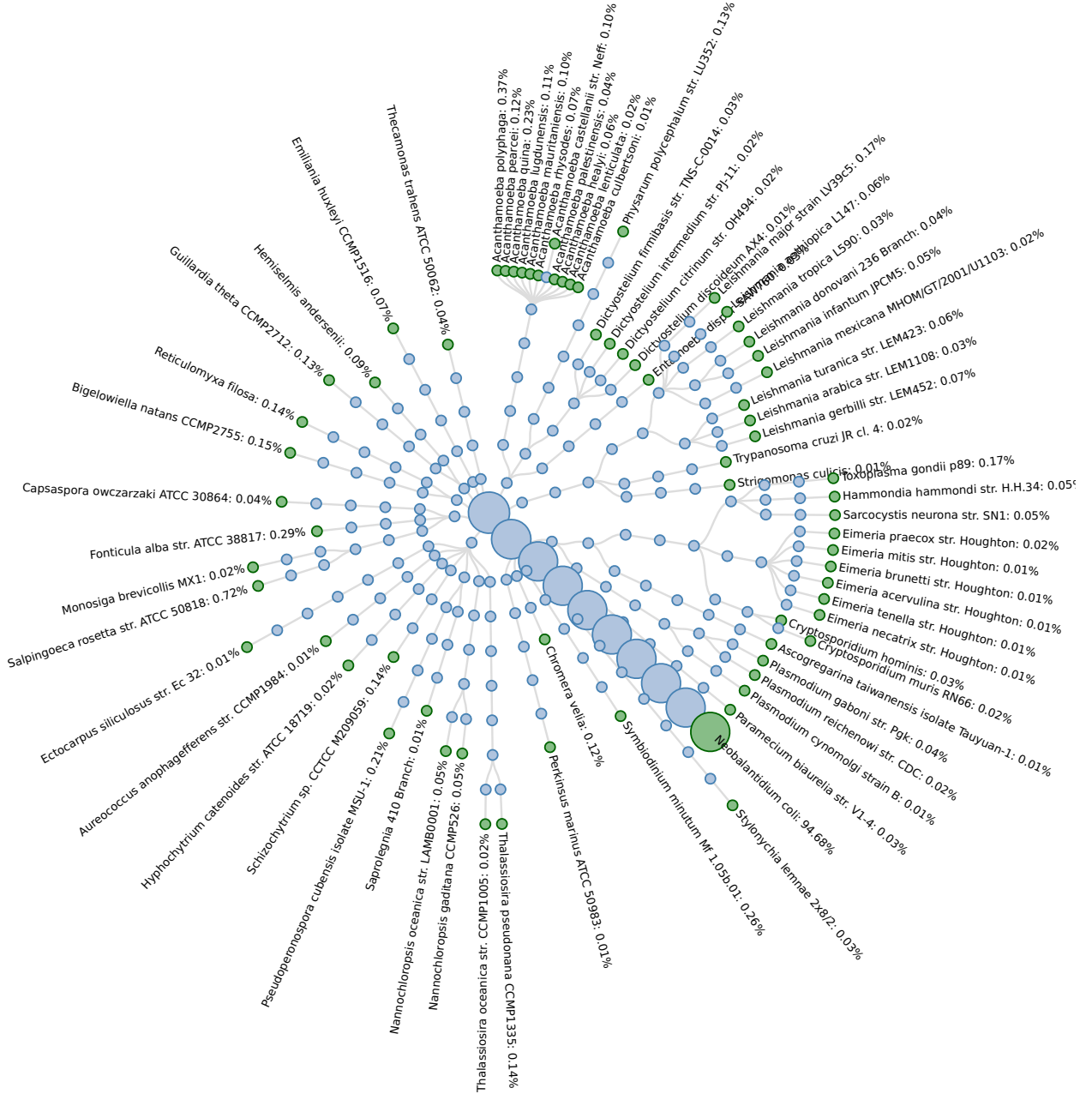


Protists

Radial Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results



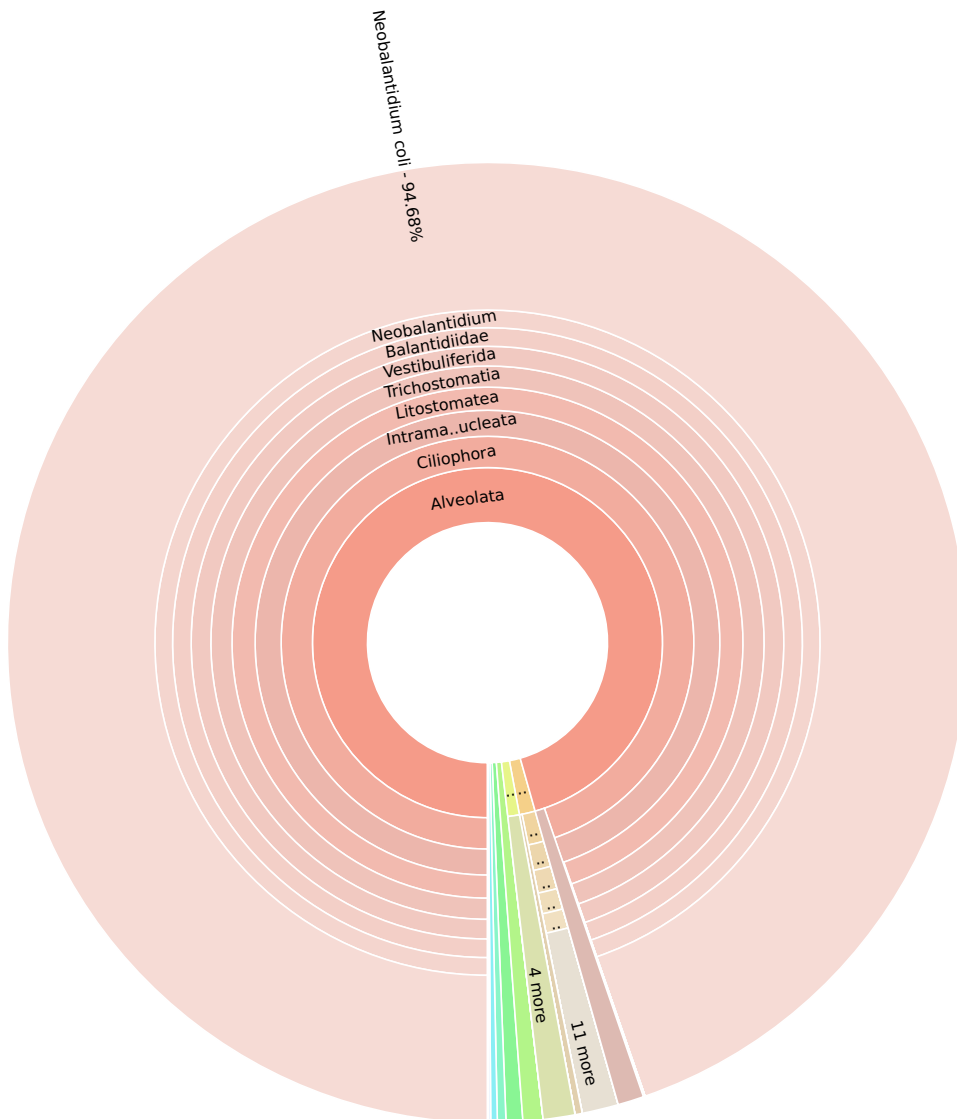


Protists

Sunburst Chart Taxonomy

Sample\_1\_BWT2214

Total results





## Dark Matter (Beta)

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	0.291M

Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Methylocystaceae bacterium UBA4761	1952232	6846.25	21.90	52.14	64.11	5548
Methylocystaceae bacterium UBA5192	1952233	6550.31	20.95	48.35	62.36	8032
Burkholderiales bacterium UBA954	1950782	3834.53	12.26	29.44	29.44	27897
Pelagibacterales bacterium UBA4760	1951182	2537.62	8.12	25.94	25.94	3073
Pseudanabaena sp.	1153	1350.87	4.32	11.14	11.23	14074
Methylocystaceae bacterium UBA2095	1952231	1311.03	4.19	10.10	39.23	4435
Burkholderiales bacterium UBA2470	1950772	946.86	3.03	9.97	11.23	5116
Burkholderiales bacterium UBA2647	1950774	721.53	2.31	7.16	11.16	1098
Lautropia sp.	2053568	550.99	1.76	6.55	11.24	1532
Pseudomonas sp. UBA6719	1947335	413.94	1.32	4.06	4.06	5252
Planctomycetaceae bacterium	2026779	313.75	1.00	2.99	2.99	7971
Novosphingobium sp. UBA6722	1946985	311.17	1.00	3.04	3.04	4356
unclassified Burkholderiales (miscellaneous)	80841	274.58	0.88	0.26	0.26	380
uncultured Collinsella sp.	165190	271.02	0.87	0.42	0.42	789
Methylophilaceae bacterium	2030816	227.36	0.73	2.88	3.50	90
Flavobacterium sp. UBA6203	1946555	224.23	0.72	2.52	2.82	802
Actinobacteria bacterium UBA3066	1948336	214.34	0.69	3.97	4.14	343
Actinobacteria bacterium	1883427	201.20	0.64	2.46	3.32	500
Burkholderiales bacterium UBA4657	1950779	173.93	0.56	1.92	1.92	2277
Chthonomonas sp. UBA2785	1946339	157.46	0.50	2.65	1.85	16
Anabaena sp. UBA12330	2055756	148.91	0.48	1.64	1.64	1756
Actinobacteria bacterium UBA2236	1948325	138.38	0.44	2.86	3.87	181
Rheinheimera sp.	1869214	136.91	0.44	1.45	1.45	2923
Polynucleobacter sp. UBA2464	1947076	128.29	0.41	0.94	0.51	750
Verrucomicrobia bacterium UBA3063	1948252	125.40	0.40	1.20	1.38	1002
unclassified Rhodanobacteraceae	1850978	121.97	0.39	2.43	2.43	28
Sediminibacterium sp. UBA657	1947456	98.19	0.31	1.38	1.38	504
Comamonadaceae bacterium UBA2334	1951601	87.88	0.28	0.94	0.94	2454
uncultured Lachnobacterium sp.	1339173	83.78	0.27	0.20	0.19	991
Synechococcales bacterium UBA8647	2055797	82.80	0.27	0.81	0.44	266
Cytophagaceae bacterium UBA6716	1951634	74.49	0.24	1.89	1.50	44
unclassified Dietzia	2617939	72.42	0.23	1.47	1.47	35
Verrucomicrobia bacterium UBA2644	1948246	68.12	0.22	1.41	1.25	211
Hylemonella sp. UBA6679	1946611	66.82	0.21	0.64	0.64	1677
Synechococcus sp. UBA8071	2055764	66.70	0.21	0.61	0.61	921
Cyanobacteria bacterium UBA6965	1947892	66.26	0.21	0.83	0.43	187
Planctomycetaceae bacterium UBA4655	1952734	62.60	0.20	0.68	0.69	2086
Mycobacterium sp.	1785	56.45	0.18	0.54	0.54	1712
Synechococcales bacterium UBA10510	2055795	53.36	0.17	0.95	0.86	155
Chloroflexi bacterium	2026724	52.89	0.17	0.72	0.72	516
Xanthomonadaceae bacterium	1926873	50.80	0.16	0.62	0.62	929
Proteobacteria bacterium UBA964	1948225	50.29	0.16	0.57	0.57	1250
Curvibacter sp.	1888168	49.84	0.16	0.48	0.50	1665
Verrucomicrobia bacterium UBA2460	1948245	49.23	0.16	1.33	1.14	91
Cytophagaceae bacterium UBA7467	1951636	47.35	0.15	1.14	1.44	128
Comamonadaceae bacterium UBA2237	1951600	40.71	0.13	0.32	0.32	900
Chitinophagaceae bacterium	1869212	39.29	0.13	0.68	0.70	130

Chitinophagaceae bacterium UBA2467	1951550	39.19	0.13	0.53	0.66	168
unclassified Citromicrobium	2630544	38.43	0.12	0.80	0.80	23
Hydrogenophaga sp.	1904254	37.28	0.12	0.61	0.38	135
Comamonas	283	35.61	0.11	0.34	0.34	279
Alphaproteobacteria bacterium UBA2784	1948371	34.88	0.11	0.48	0.48	642
Comamonadaceae bacterium	1871071	34.77	0.11	0.40	0.40	1010
Acidovorax sp.	1872122	33.15	0.11	0.36	0.38	1337
Legionellales bacterium	2026754	31.18	0.10	0.54	0.54	163
Ramlibacter tataouinensis	94132	30.84	0.10	0.21	0.21	843
bacterium UBA951	1947788	31.03	0.10	0.60	0.60	93
Cyanobacteria bacterium UBA7373	1947895	29.63	0.10	0.32	0.32	520
Candidatus Microthrix	41949	29.70	0.10	1.15	1.15	5
Hyphomonadaceae bacterium	2026748	28.42	0.09	0.37	0.37	657
Prochlorococcus sp. UBA3999	1947246	27.14	0.09	0.44	0.34	102
Parachlamydiales bacterium	2052178	27.24	0.09	0.29	0.29	377
Proteobacteria bacterium UBA4656	1948178	26.61	0.08	0.33	0.33	957
Streptomyces sp.	1931	26.09	0.08	0.79	0.79	13
Comamonadaceae bacterium UBA6202	1951611	24.91	0.08	0.29	0.29	798
Synechococcales bacterium UBA12195	2055798	24.39	0.08	0.39	0.35	65
Acidimicrobium sp.	1872112	24.52	0.08	0.42	0.50	132
Tepidimonas sp. UBA997	1947650	23.88	0.08	0.29	0.29	582
Polaromonas sp.	1869339	23.67	0.08	0.19	0.13	141
unclassified Bradyrhizobium	2631580	23.45	0.07	0.30	0.30	133
unclassified Comamonadaceae	83494	23.33	0.07	0.19	0.19	248
Rhodoferax sp. UBA4409	1947378	22.35	0.07	0.23	0.24	768
Pseudomonas stutzeri	316	21.92	0.07	0.29	0.28	90
Rhodobacter sp.	1062	21.47	0.07	0.24	0.24	725
Aeromonas veronii	654	21.70	0.07	0.29	0.31	505
Achromobacter	222	21.41	0.07	0.16	0.16	61
Rhodocyclaceae bacterium UBA2250	1952837	20.28	0.07	0.27	0.27	521
Ralstonia	48736	19.90	0.06	0.30	0.30	15
Armatimonadetes bacterium	2033014	20.02	0.06	0.65	1.78	18
Comamonadaceae bacterium UBA4962	1951608	19.78	0.06	0.23	0.24	846
Alicyclophilius sp. UBA7619	1946005	19.05	0.06	0.24	0.26	755
Legionellales bacterium UBA4759	1951149	19.10	0.06	0.37	0.37	110
Comamonas aquatica	225991	18.10	0.06	0.23	0.25	520
unclassified Legionellales (miscellaneous)	2570685	17.97	0.06	0.10	0.10	43
Aquabacterium sp. UBA2148	1946042	16.88	0.05	0.23	0.31	312
Acinetobacter sp. UBA4611	1945942	16.82	0.05	0.39	0.61	55
Acidovorax delafieldii	47920	16.44	0.05	0.21	0.23	609
Acidimicrobiaceae bacterium	2024894	15.93	0.05	0.20	0.20	685
Rhodobacteraceae bacterium UBA1943	1952798	15.17	0.05	0.17	0.17	570
Candidatus Accumulibacter	327159	15.22	0.05	0.11	0.08	92
Mycobacterium tuberculosis	1773	15.24	0.05	0.15	0.15	876
Micrococcus luteus	1270	15.34	0.05	0.13	0.13	41
Verrucomicrobiaceae bacterium UBA2429	1952942	15.41	0.05	0.28	0.22	97
Plesiomonas shigelloides	703	14.96	0.05	0.25	0.33	212
Burkholderiales bacterium UBA1834	1950768	14.68	0.05	0.25	0.21	163
unclassified Gordonia	2657482	14.45	0.05	0.14	0.14	207
Porphyrobacter sp. UBA7686	1947080	14.09	0.04	0.22	0.22	275
Dechloromonas sp. UBA6271	1946404	14.18	0.04	0.19	0.20	478
Thiobacillus denitrificans	36861	13.88	0.04	0.32	0.17	47
Burkholderiales bacterium UBA4200	1950777	13.41	0.04	0.29	0.21	65
bacterium UBA7470	1947819	13.29	0.04	0.19	0.19	336
Castellaniella sp. UBA6218	1946233	11.77	0.04	0.17	0.17	342

Verrucomicrobia bacterium UBA3062	1948251	11.77	0.04	0.17	0.17	204
unclassified Sphingopyxis	2614943	11.43	0.04	0.50	0.50	5
Gemmobacter sp.	1898957	11.45	0.04	0.21	0.13	76
Betaproteobacteria bacterium UBA5582	1948460	11.60	0.04	0.14	0.16	447
Verrucomicrobiales bacterium	2026801	11.63	0.04	0.16	0.16	498
Nitrosomonas sp. UBA6494	1946979	11.32	0.04	0.19	0.19	190
Blastomonas sp. UBA7677	1946117	11.02	0.03	0.19	0.19	198
Massilia sp.	1882437	11.09	0.03	0.17	0.17	34
Syntrophaceae bacterium UBA5744	1961444	10.70	0.03	0.15	0.17	366
Acinetobacter	469	10.24	0.03	0.44	0.95	2
Caulobacteraceae bacterium UBA3198	1951537	9.85	0.03	0.37	0.21	55
Comamonadaceae bacterium UBA3515	1951605	9.97	0.03	0.10	0.13	425
Gammaproteobacteria bacterium	1913989	9.99	0.03	0.21	0.11	43
Runella sp.	1960881	9.86	0.03	0.16	0.21	207
Hyphomonadaceae bacterium UBA7672	1951957	9.57	0.03	0.14	0.14	382
Candidatus Accumulibacter phosphatis	327160	9.63	0.03	0.26	0.15	31
unclassified Brevundimonas	2622653	9.34	0.03	0.12	0.12	25
Rhodobacteraceae bacterium UBA6273	1952813	9.28	0.03	0.13	0.13	331
Rhodobacteraceae bacterium UBA6197	1952810	9.24	0.03	0.12	0.12	362
Acidimicrobiaceae bacterium UBA668	1951340	9.49	0.03	0.16	0.17	156
Gemmatimonas	173479	9.33	0.03	10.26	10.26	7
Comamonas sp. UBA7840	1946392	9.02	0.03	0.13	0.14	253
Competibacteraceae bacterium UBA3908	1951621	8.97	0.03	0.13	0.13	327
unclassified Thauera	2609274	8.66	0.03	0.12	0.08	29
Micrococcales bacterium UBA7469	1951173	8.67	0.03	0.16	0.16	97
Rhodobacteraceae bacterium UBA6796	1952814	8.01	0.03	0.11	0.11	283
Delftia acidovorans	80866	8.10	0.03	0.12	0.21	186
Delftia lacustris	558537	8.26	0.03	0.12	0.21	277
Thauera sp. UBA6194	1947679	8.24	0.03	0.12	0.13	2977
unclassified Actinomycetales (miscellaneous)	105426	8.02	0.03	0.39	0.39	3
Thiomonas sp. UBA7699	1947694	7.94	0.03	0.11	0.11	273
Thiobacillus sp. UBA2597	1947687	7.78	0.03	0.12	0.12	183
Arenimonas sp.	1872635	7.78	0.03	0.12	0.12	233
unclassified Exiguobacterium	2644629	7.82	0.03	0.15	0.17	38
uncultured Clostridium sp.	59620	7.96	0.03	0.12	0.12	42
unclassified Cytophagaceae	94253	7.82	0.03	0.11	0.13	88
Actinobacteria bacterium UBA3007	1948330	7.46	0.02	0.21	0.21	29
Rhodocyceae bacterium UBA4043	1952842	7.28	0.02	0.11	0.12	229
Betaproteobacteria bacterium UBA7395	1948463	7.05	0.02	0.11	0.14	187
Chitinophagaceae bacterium UBA3961	1951555	7.14	0.02	0.14	0.14	121
Actinobacteria bacterium UBA5975	1948344	7.02	0.02	0.11	0.11	103
Planctomycetes bacterium UBA2386	1948127	6.90	0.02	0.11	0.11	290
Opitutae bacterium UBA953	1948744	6.10	0.02	0.10	0.10	171
unclassified Proteobacteria (miscellaneous)	81684	6.06	0.02	0.11	0.09	2
Verrucomicrobiaceae bacterium UBA2021	1952940	5.89	0.02	0.19	0.24	17
unclassified Aminicenantes	910038	5.79	0.02	0.19	0.19	11
uncultured Prevotella sp.	159272	5.58	0.02	0.16	0.11	15
Acidobacteria bacterium	1978231	5.67	0.02	0.15	0.15	32
Proteobacteria bacterium UBA2646	1948154	5.27	0.02	0.16	0.14	19
Synechococcales bacterium UBA8138	2055796	5.35	0.02	0.10	0.10	406
unclassified Acidimicrobiaceae	667305	5.18	0.02	0.19	0.19	9
unclassified Flavobacteriales (miscellaneous)	403978	5.19	0.02	3.23	3.23	9
Crocinitomicaceae bacterium	2026728	4.86	0.02	0.10	0.10	64
unclassified Pseudomonas	2583993	4.57	0.01	0.28	0.28	3
Pseudomonas kunmingensis	1211807	4.26	0.01	0.16	0.14	12

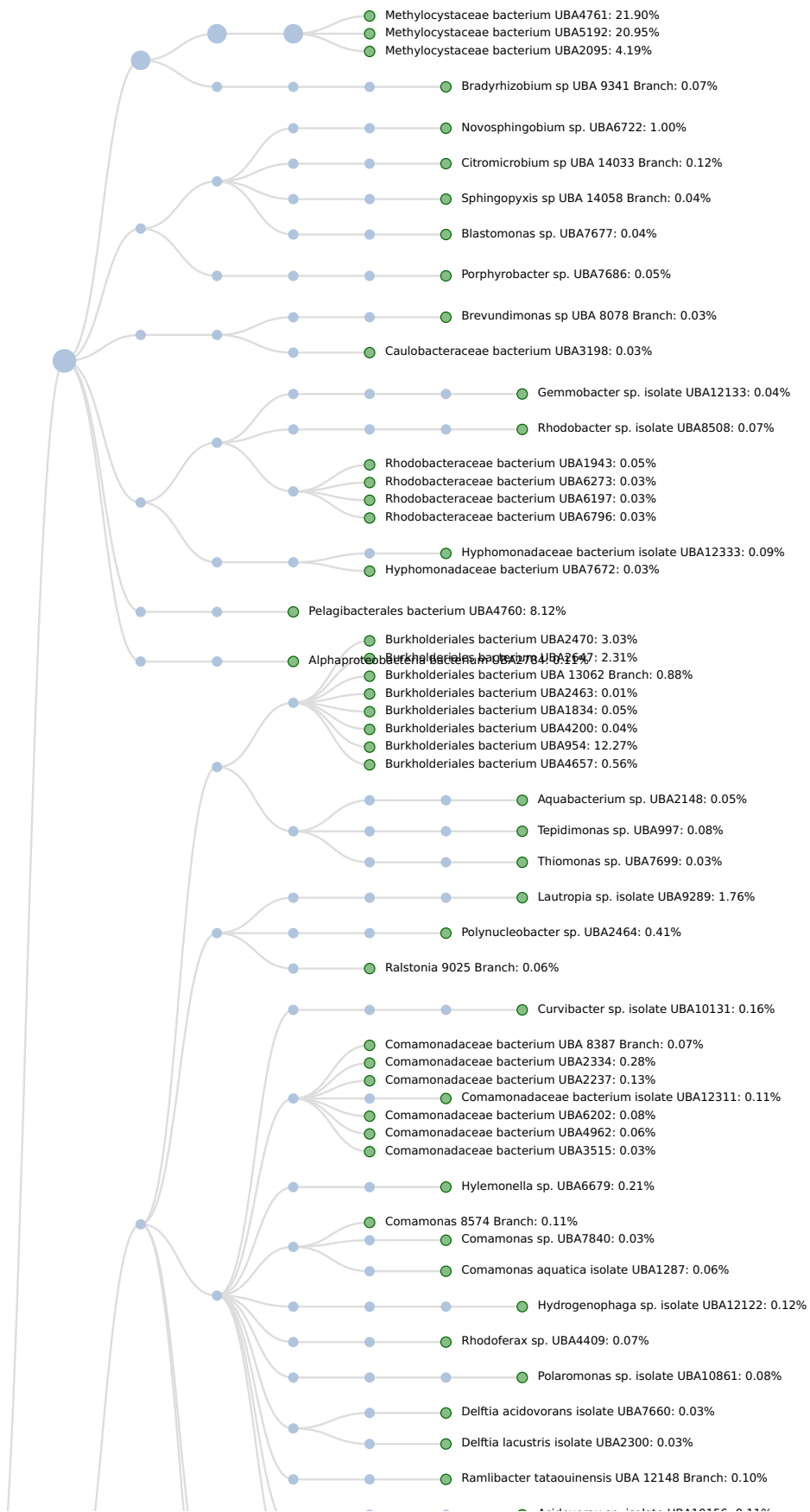
Burkholderiales bacterium UBA2463	1950771	4.11	0.01	0.37	0.27	15
unclassified Cellvibrio	2624793	3.62	0.01	4.96	4.96	11
unclassified Patescibacteria group	1948766	3.87	0.01	0.25	0.25	5
Francisellaceae bacterium UBA6186	1951894	2.37	<0.01	0.10	0.10	18
Shigella sp.	625	1.90	<0.01	0.34	0.34	7
Flavobacteriales bacterium UBA6717	1951053	1.46	<0.01	0.17	0.12	6

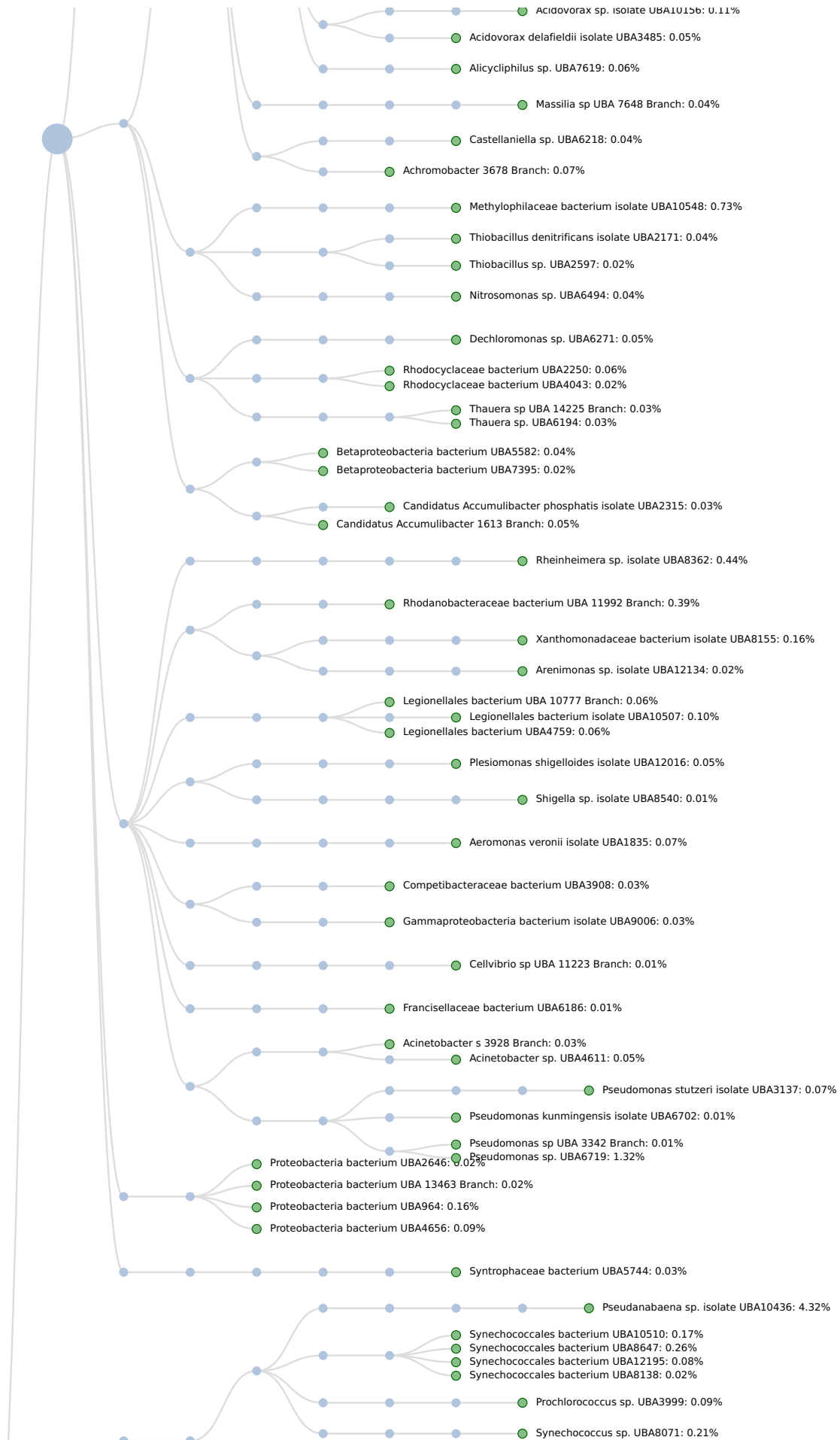
Dark Matter (Beta)

Tree Chart Taxonomy

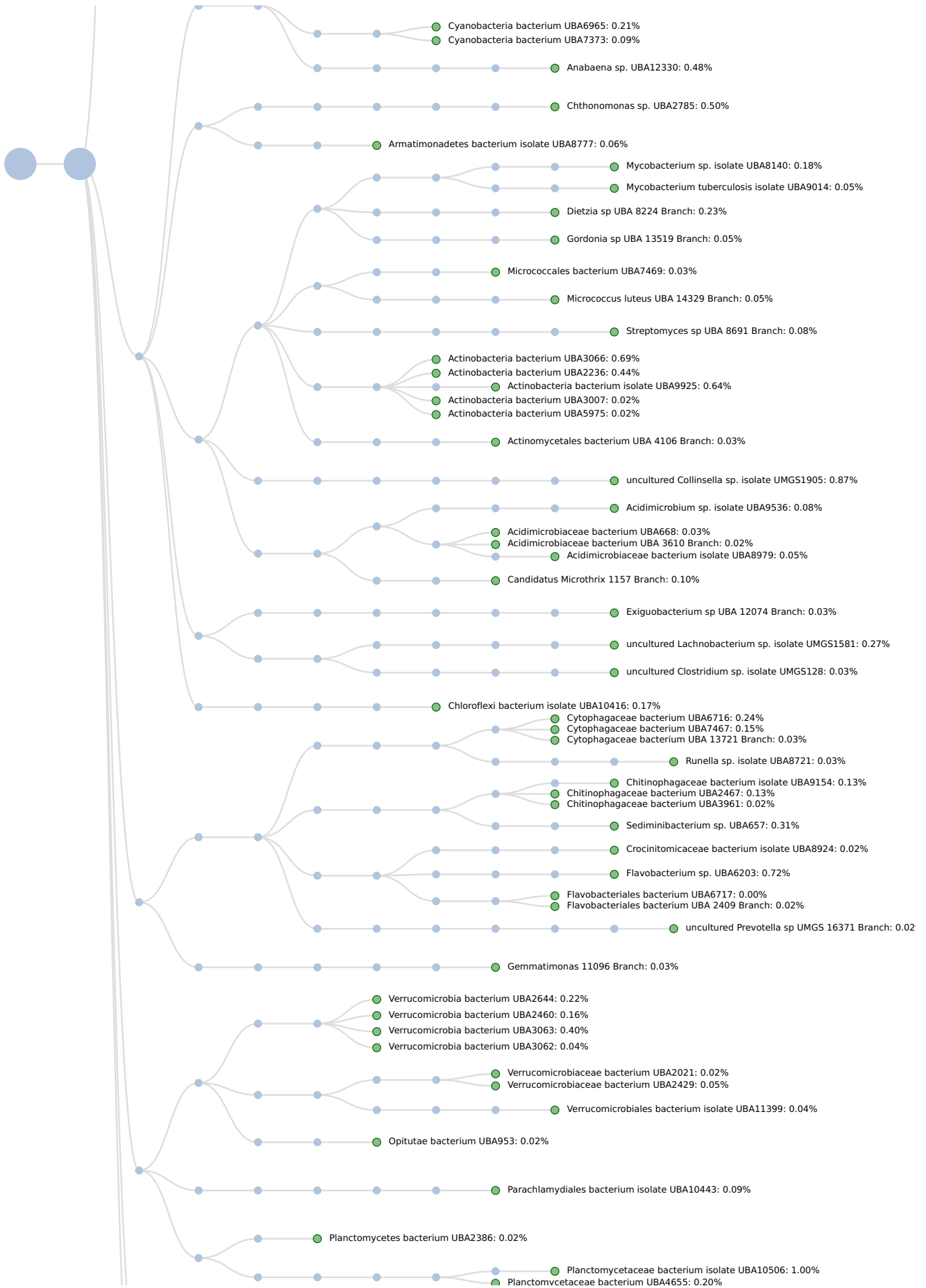
Sample\_1\_BWT2214

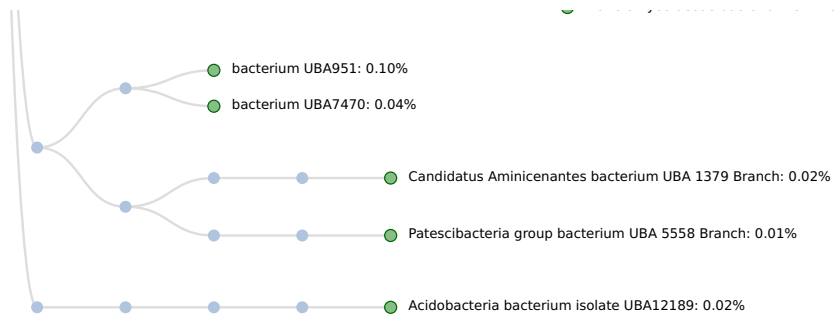
Total results









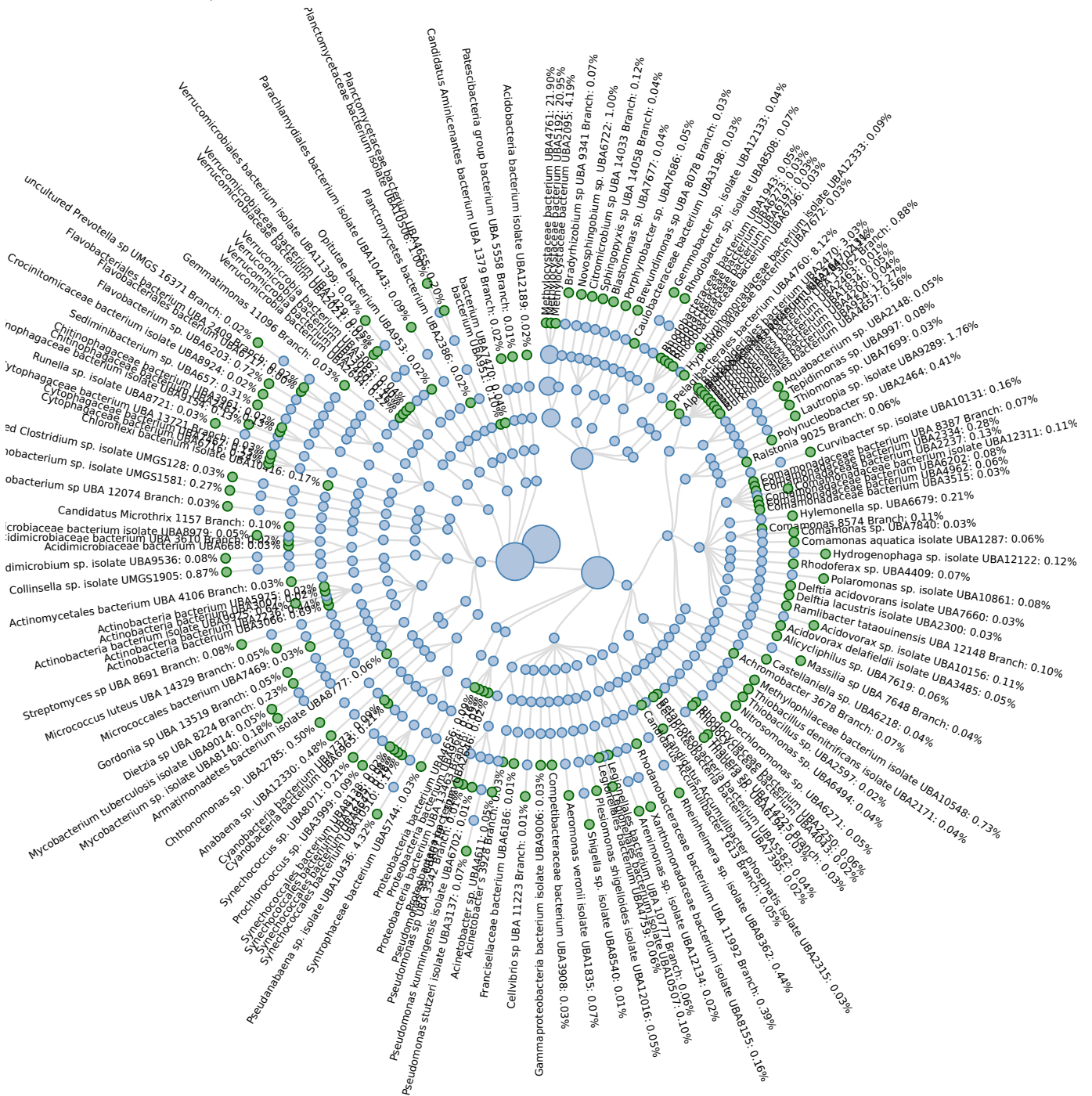


# Dark Matter (Beta)

Radial Tree Chart Taxonomy

# Sample\_1\_BWT2214

Total results



Dark Matter (Beta)

Sunburst Chart Taxonomy

Sample\_1\_BWT2214

Total results

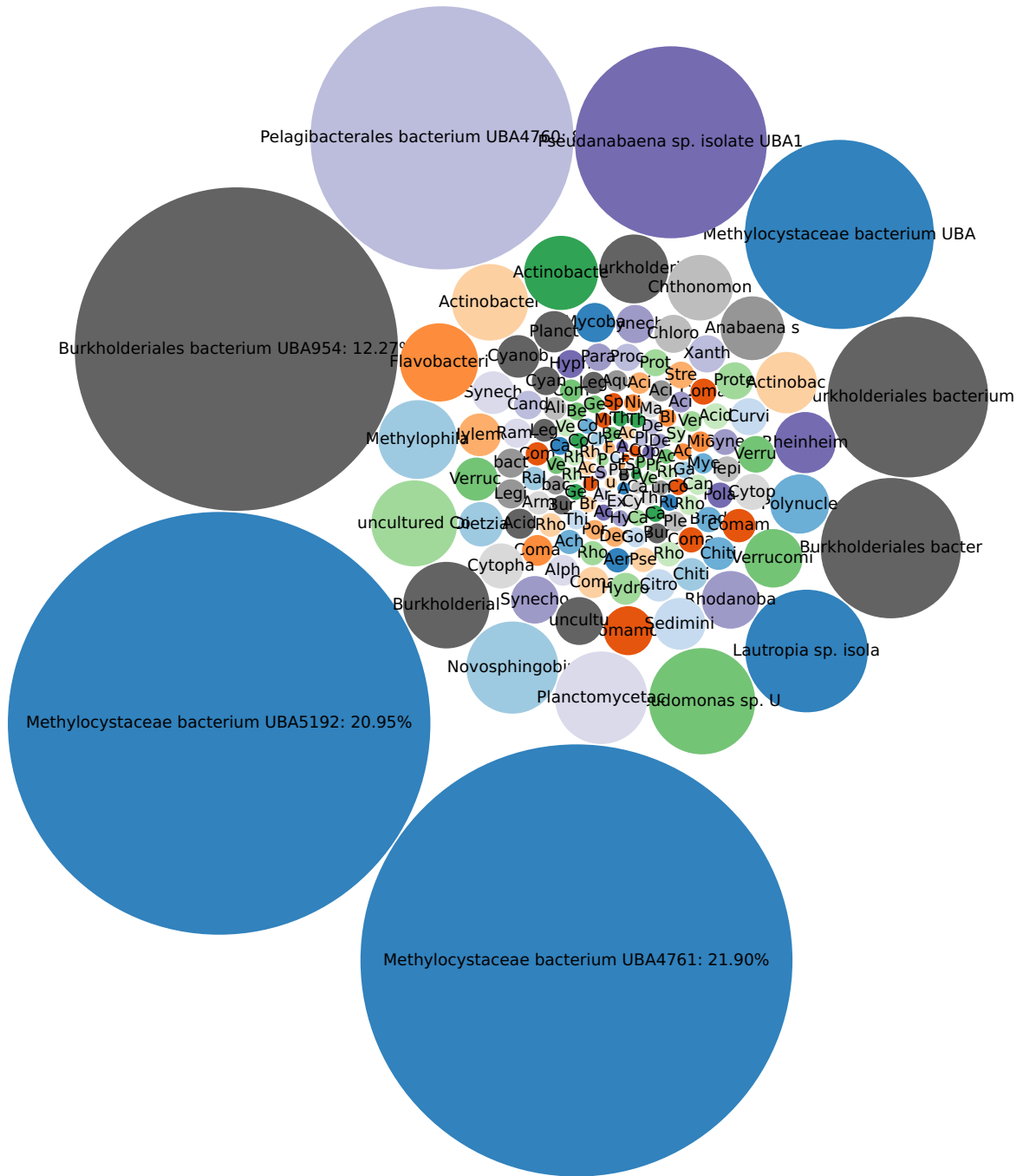


Dark Matter (Beta)

Bubble Chart Taxonomy

Sample\_1\_BWT2214

Total results



## Phages

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	84.508k

### Table

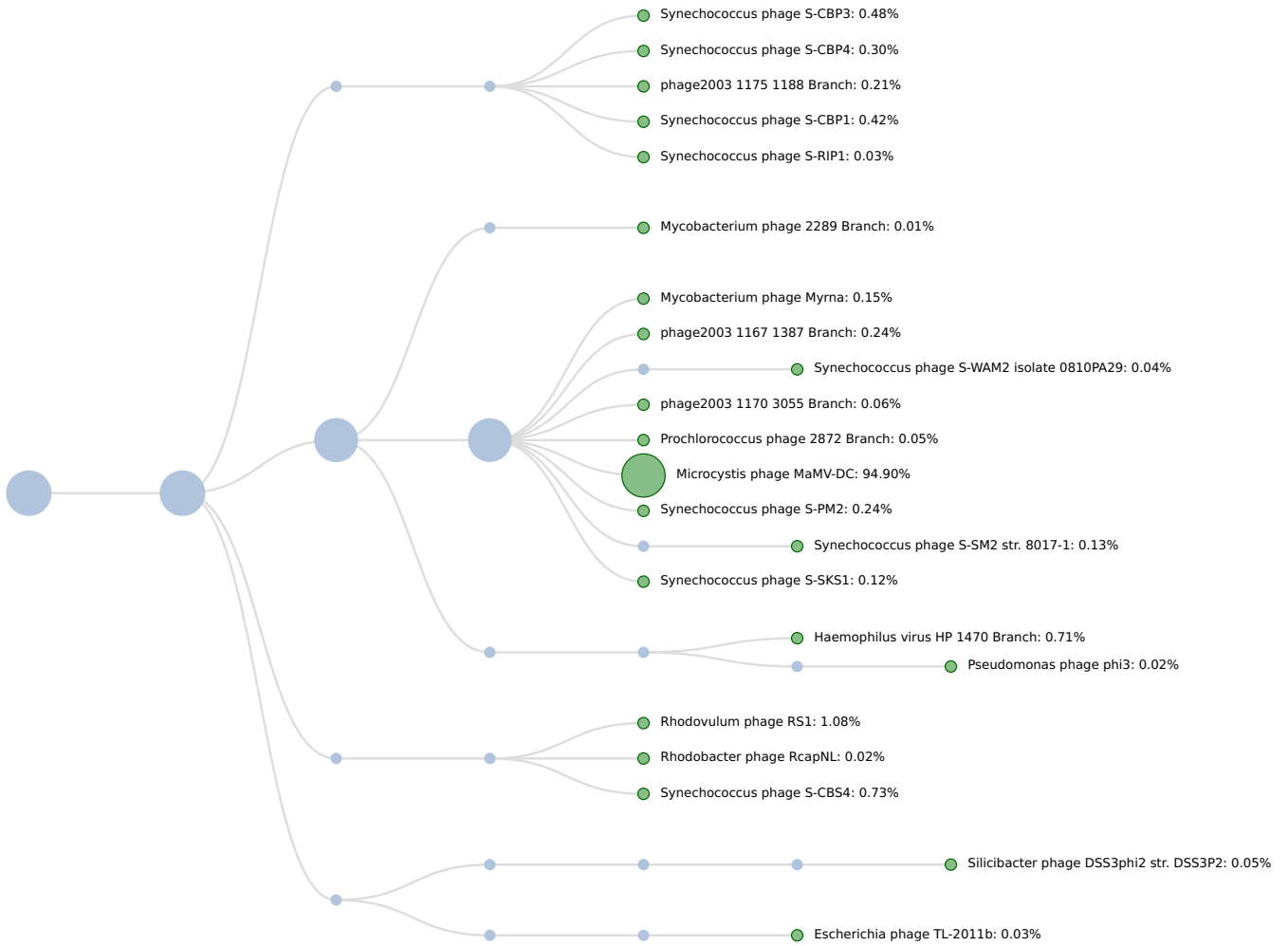
Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis phage MaMV-DC	1357715	13989.42	94.90	36.25	36.25	302583
Rhodovulum phage RS1	754056	158.77	1.08	1.92	1.92	58
Synechococcus phage S-CBS4	756275	107.06	0.73	0.69	0.69	1670
Hpunavirus	1196844	104.94	0.71	0.24	0.24	655
Synechococcus phage S-CBP3	756276	70.25	0.48	0.85	1.31	536
Synechococcus phage S-CBP1	1273711	61.67	0.42	0.60	0.95	619
Synechococcus phage S-CBP4	754059	44.19	0.30	0.71	1.08	254
unclassified Myoviridae	196896	35.67	0.24	0.40	0.40	161
Synechococcus phage S-PM2	238854	35.62	0.24	0.37	0.39	922
unclassified Autographiviridae	2731990	30.69	0.21	0.23	0.23	433
Mycobacterium phage Myrna	546805	21.98	0.15	0.11	0.12	809
Synechococcus phage S-SM2	444860	18.80	0.13	0.26	0.30	428
Synechococcus phage S-SKS1	754042	17.16	0.12	0.25	0.29	354
unclassified Myoviridae	196896	9.29	0.06	0.11	0.11	20
Silicibacter phage DSS3phi2	490912	7.76	0.05	0.23	0.22	68
unclassified Myoviridae	196896	7.53	0.05	0.15	0.15	22
Synechococcus phage S-WAM2	1815522	6.33	0.04	0.10	0.20	176
Escherichia phage TL-2011b	1124654	4.50	0.03	0.17	0.14	25
Synechococcus phage S-RIP1	754041	3.92	0.03	0.11	0.12	32
Pseudomonas phage phi3	1754217	2.72	0.02	0.13	0.13	51
Rhodobacter phage RcapNL	1131316	2.27	0.01	0.11	0.11	214
Bixzunavirus	680114	1.38	<0.01	0.12	0.17	304

Phages

Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results

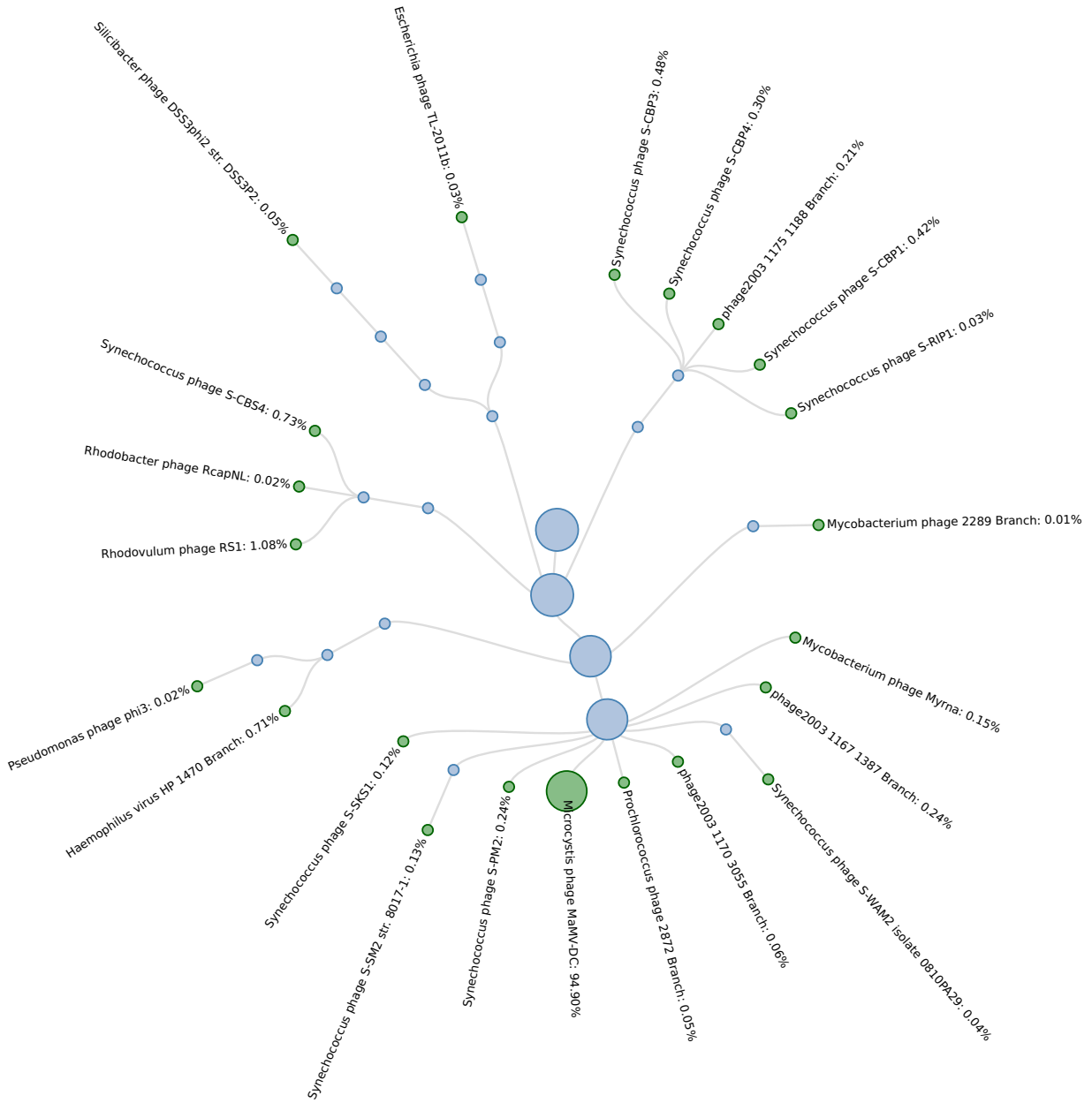


Phages

Radial Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results



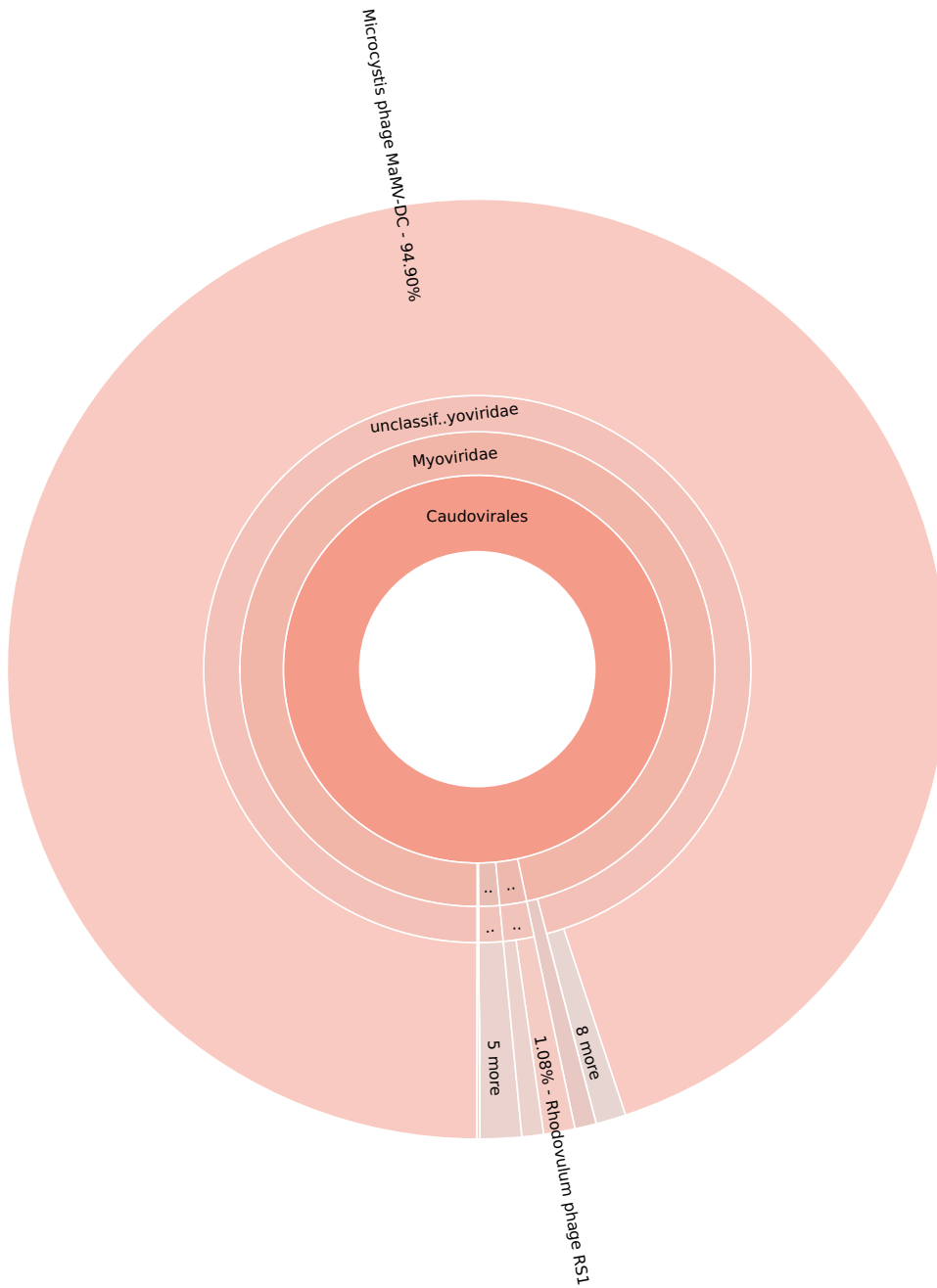


Phages

Sunburst Chart Taxonomy

Sample\_1\_BWT2214

Total results





## Viruses

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	0.172M

### Table

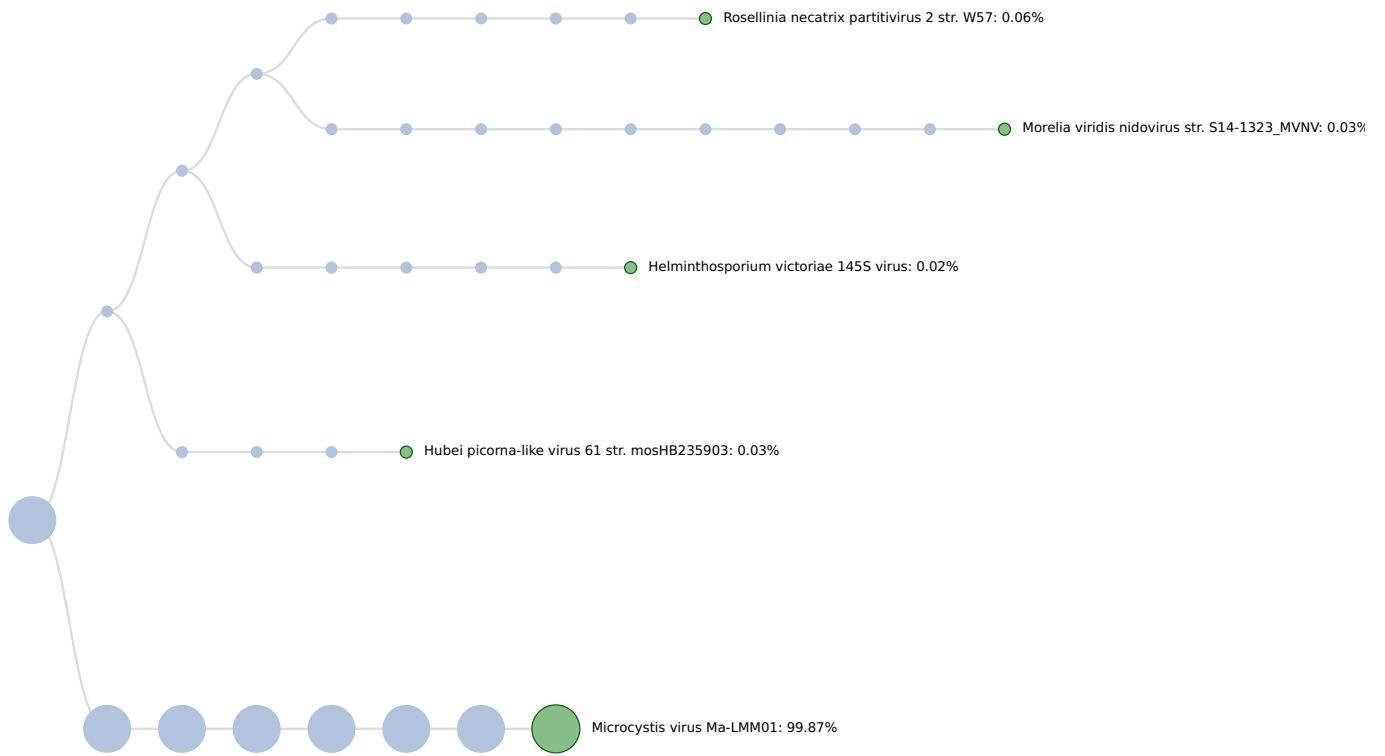
Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis virus Ma-LMM01	340435	15685.16	99.87	40.51	40.51	336749
Rosellinia necatrix partitivirus 2	859651	9.19	0.06	0.52	0.52	104
Hubei picorna-like virus 61	1923144	4.24	0.03	0.24	0.24	97
Morelia viridis nidovirus	2016400	4.07	0.03	0.16	0.16	266
Helminthosporium victoriae 145S virus	164750	2.71	0.02	0.15	0.15	171

# Viruses

Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results

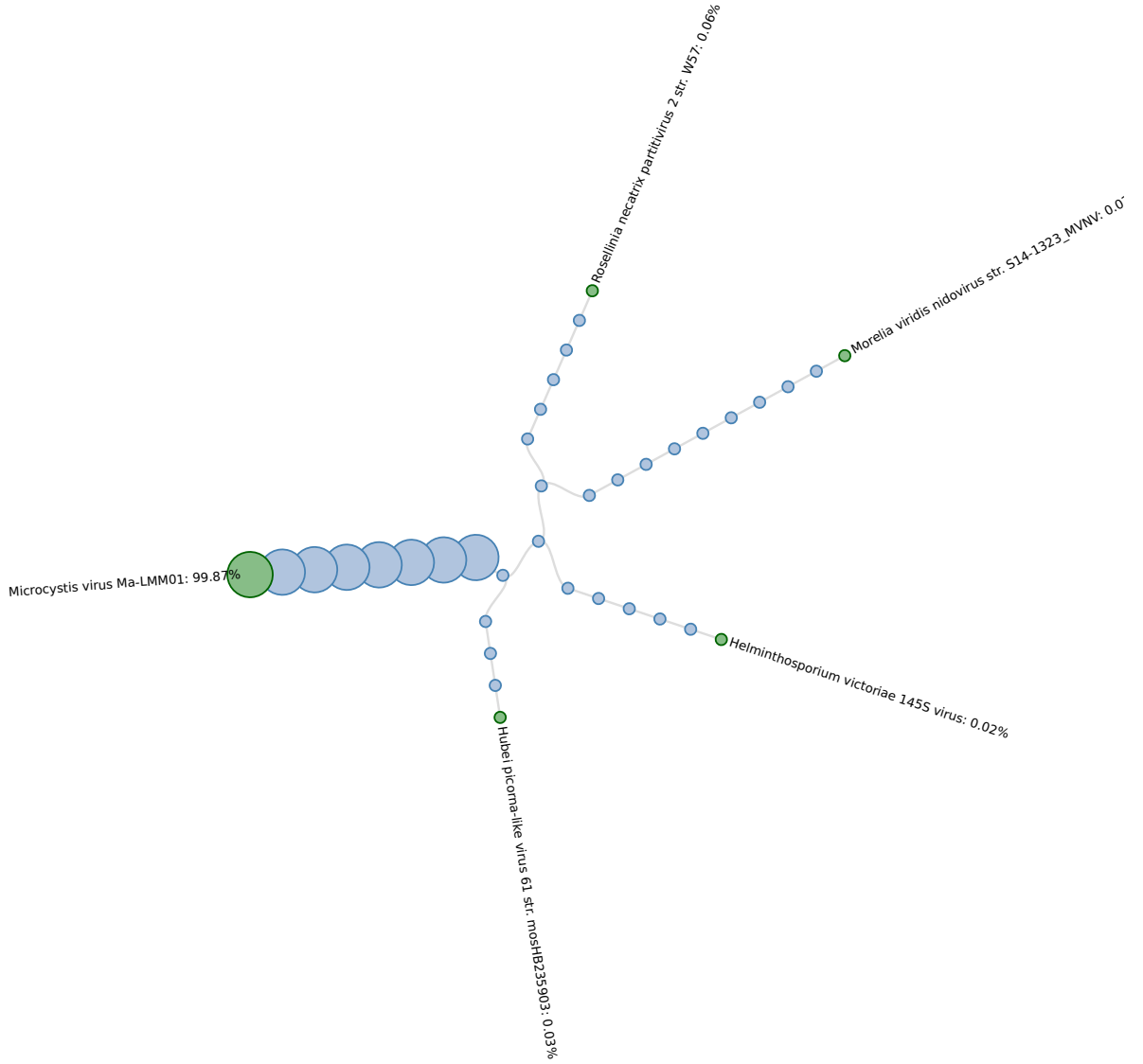


Viruses

Radial Tree Chart Taxonomy

Sample\_1\_BWT2214

Total results

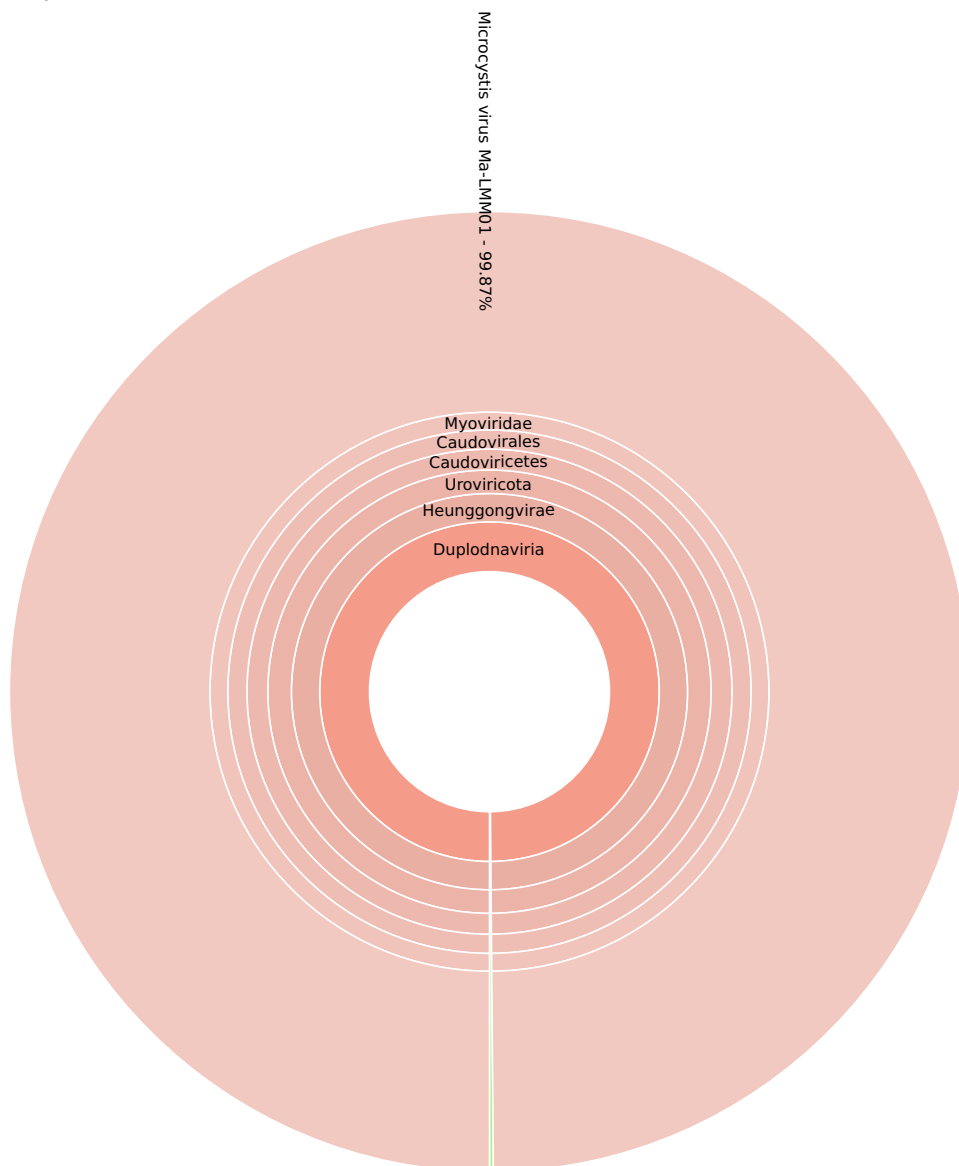


# Viruses

Sunburst Chart Taxonomy

Sample\_1\_BWT2214

Total results

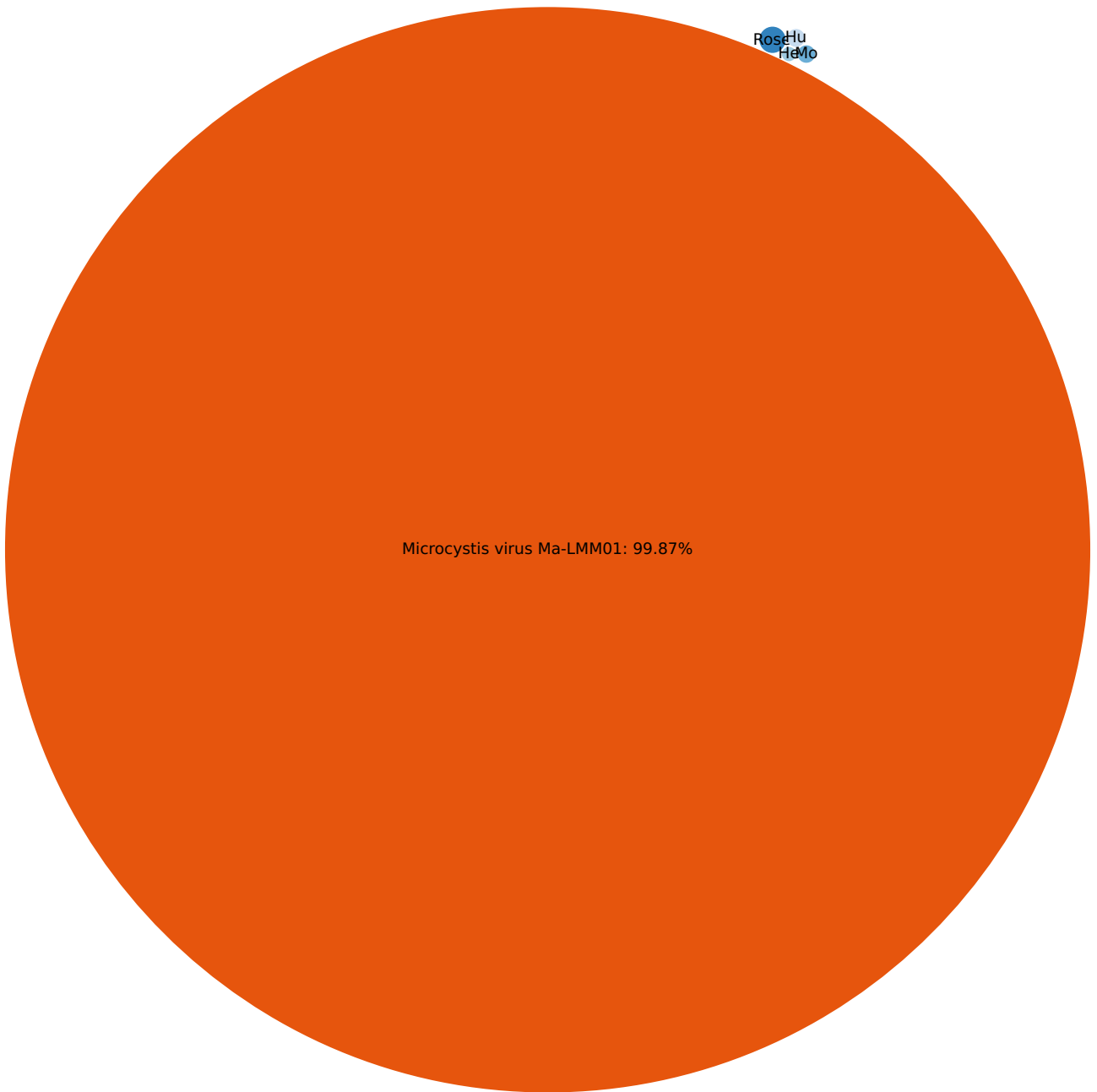


Viruses

Bubble Chart Taxonomy

Sample\_1\_BWT2214

Total results



## Respiratory Virus

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	7.319k

Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Human mastadenovirus C	129951	25.50	100.00	0.14	0.08	117



## Virulence Factors

Sample\_1\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	2.844k

### Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Pseudomonas aeruginosa GENE intI1	-	7301.06	32.65	98.00	98.00	201
Vibrio cholerae GENE intI1	-	6824.83	30.52	100.00	100.00	76
Enterobacter aerogenes GENE tniC	-	5644.85	25.25	77.78	77.78	372
Mycobacterium tuberculosis GENE icl-aceA	-	1119.85	5.01	12.26	12.26	233
Enterobacter aerogenes GI 1572570	-	892.94	3.99	12.74	12.74	177
Pseudomonas aeruginosa GENE xcpT	-	576.65	2.58	14.29	14.29	37

## Disclaimer

CosmosID is designed for Research Use Only in accordance with applicable rules and regulations of the United States Food and Drug Administration and other applicable laws, and your sample shall not be used for patient care or diagnostic, clinical or therapeutic use. You agree to use CosmosID for Research Use Only. No analyses, reports, or other information obtained or provided through CosmosID are intended to be (nor should be relied upon as) medical advice or instructions for medical diagnosis or treatment.

CosmosID does not accept any responsibility for the accuracy of the input entered by the user or the consequences of any inaccuracies in this input. The analyses are not intended to replace professional medical care and attention by a qualified medical practitioner and consequently CosmosID does not accept any responsibility for the selection of drugs and the patient's response to treatment.

CosmosID is designed for whole genome shotgun (WGS) analysis on reads of at least 75 bases in length and should not be used for 16S analysis.

# Appendix D2



## CosmosID - Unlocking the Microbiome

Filename:	Sample_2_BWT2214
Size:	10.35 GiB
Reads:	51.468M
Date of Report:	2021-08-15 00:46:36

## Summary

Database Name	Hits	Status
Bacteria	172	Success
Antibiotic Resistance	0*	Success
Fungi	18	Success
Protists	15	Success
Dark Matter (Beta)	153	Success
Phages	21	Success
Viruses	2	Success
Respiratory Virus	0*	Success
Virulence Factors	8	Success

\* Analysis with 0 hits will not be included in this report

## Bacteria

Sample\_2\_BWT2214

## Fasta/q details

Total results

Metric	Value
Hit	7.771M

## Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis panniformis FACHB-1757	1638788	547502.38	57.96	75.70	86.05	1727691
Microcystis aeruginosa PCC 9807	1160283	177248.94	18.76	43.04	67.64	306982
Flavobacterium fontis	1124188	92874.64	9.83	41.84	42.00	2046337
Microcystis sp. 0824	1502726	47987.34	5.08	23.13	37.64	96107
Microcystis viridis NIES-102	213615	42672.82	4.52	24.16	42.54	36600
Dolichospermum circinale AWQC131C	398007	5648.07	0.60	33.07	43.24	33261
Aquidulcibacter paucihalophilus	1978549	4948.80	0.52	14.12	14.12	139696
Clavibacter sp.	1871044	4433.20	0.47	33.70	33.70	11827
Silanimonas lenta DSM 16282	1123253	2705.85	0.29	2.63	2.63	76650
Flavobacterium sp. WWJ-16	2506421	1766.53	0.19	2.18	2.97	35424
Novosphingobium ginsenosidimutans	1176536	1483.49	0.16	9.10	9.10	29266
Candidatus Fonsibacter ubiqus	1925548	1314.99	0.14	14.70	14.72	2079
Flectobacillus sp. BAB-3569	1509483	891.95	0.09	7.59	7.61	16423
Synechococcus sp. CB0101	232348	825.08	0.09	6.68	6.78	10829
Inhella crocodyli	2499851	795.59	0.08	6.80	6.80	17603
Pseudanabaena sp. ABRG5-3	685565	786.46	0.08	1.80	1.84	21934
Erythrobacter neustonensis	1112	784.84	0.08	7.21	7.21	11188
Leptospira ryugeni	1917863	769.92	0.08	7.55	7.57	6649
Phenylobacterium parvum	2201350	690.49	0.07	4.23	4.23	16450
Pararheinheimera texasensis DSM 17496	1123055	633.13	0.07	5.51	5.51	12035
Tabrizicola sp. TH137	2067452	484.12	0.05	3.42	3.78	11726
Limnothrix sp. P13C2	1880902	424.27	0.04	1.84	4.02	44
Rhodobacter sp. CACIA14H1	1408890	403.60	0.04	2.68	3.05	10347
Raphidiopsis brookii D9	533247	348.84	0.04	3.02	3.86	1597
Aquimonas voraii	265719	325.58	0.03	2.69	2.69	9819
Porphyrobacter sp. LM 6	1896196	251.89	0.03	2.59	2.59	3807
Aphanizomenon flos-aquae 2012/KM1/D3	1532906	234.00	0.03	1.94	2.69	1162
Dolichospermum sp. UHCC 0315A	1914871	222.60	0.02	1.20	2.05	1067
Anabaena sp. 90	46234	207.83	0.02	1.35	2.10	850
Sphingorhabdus contaminans	1343899	173.10	0.02	1.86	1.87	3017
Aquabacterium sp. KMB7	2528630	173.09	0.02	1.82	1.82	3734
Vulcanococcus limneticus LL	2025607	148.05	0.02	1.41	1.43	2997
Pseudomonas sp. HAR-UPW-AIA-41	1985301	153.60	0.02	1.68	1.68	2641
Pseudanabaena biceps PCC 7429	927668	138.54	0.01	0.47	0.48	4499
Elstera cyanobacteriorum	2022747	142.09	0.01	1.55	1.55	3041
Sphaerotilus natans subsp. natans DSM 6575	1286631	145.96	0.01	1.49	1.49	3857
Arenimonas metalli CF5-1	1384056	109.08	0.01	0.20	0.25	5621
beta proteobacterium SCGC AAA028-K02	938797	113.23	0.01	1.52	1.52	364
Flavobacterium sasangense DSM 21067	1121896	112.08	0.01	1.47	1.59	9191
Cylindrospermopsis sp. CR12	1747196	108.12	0.01	0.68	2.15	831
Flavobacterium filum DSM 17961	1121889	99.81	0.01	0.13	0.13	2766
Candidatus Planktophila sp.	2175601	95.39	0.01	1.13	1.57	127
Ideonella sp. KYPY4	1862385	90.12	0.01	0.74	0.74	3898
Polynucleobacter cosmopolitanus	351345	96.55	0.01	1.30	1.33	627
Cyanobium sp. NIES-981	1851505	86.89	<0.01	0.94	1.20	1533
Arenimonas malthae CC-JY-1	1384054	85.95	<0.01	0.21	0.26	4489
Arenimonas composti TR7-09 = DSM 18010	1121013	83.02	<0.01	0.16	0.16	5300

Hydrogenophaga sp. IBVHS2	1985170	85.20	<0.01	0.96	0.96	1687
Cyanobium gracile PCC 6307	292564	74.49	<0.01	0.87	2.50	1191
Piscinibacter defluvi	1796922	73.89	<0.01	0.36	0.52	4665
Polynucleobacter victoriensis	2049319	77.15	<0.01	1.05	1.09	516
Aphanothece cf. minutissima CCALA 015	2107695	70.23	<0.01	0.90	2.60	964
Cyanobium usitatum str. Tous	2116684	70.33	<0.01	0.79	0.86	1047
actinobacterium SCGC AAA027-J17	932040	64.34	<0.01	1.05	1.05	184
Arenimonas caeni	2058085	62.59	<0.01	0.21	0.26	3007
Rubrivivax albus	2499835	65.74	<0.01	0.36	0.41	4005
Cylindrospermopsis raciborskii CS-505	533240	60.33	<0.01	0.89	2.27	278
Arenimonas terrae	2546226	55.07	<0.01	0.13	0.16	3333
Niveispirillum cyanobacteriorum	1612173	58.11	<0.01	0.64	0.80	1799
Cetobacterium somerae ATCC BAA-474	1319815	58.56	<0.01	0.88	0.89	247
Mycobacterium sp. UM_11	1638773	49.67	<0.01	1.85	1.85	11
Rhizobium sp. MSSRF QS100	1522278	47.36	<0.01	0.64	1.16	651
Pseudomonas alcaligenes NBRC 14159	1215092	43.76	<0.01	0.54	1.08	761
Niveispirillum lacus	1981099	44.14	<0.01	0.49	0.66	1472
Ideonella sakaiensis	1547922	49.67	<0.01	0.28	0.40	3164
Zhizhongheella caldifontis	1452508	49.43	<0.01	0.41	0.58	2027
Lysobacter tabacisoli	2315424	35.45	<0.01	0.11	0.11	2639
Caedibacter taeniospiralis	28907	38.18	<0.01	0.65	0.65	215
Sandarakinorhabdus cyanobacteriorum	1981098	39.25	<0.01	0.44	0.48	730
Erythrobacter colymbi	1161202	35.99	<0.01	0.42	0.72	1038
Caulobacter sp. CCH4-E1	1768763	35.34	<0.01	0.80	0.69	113
Brevundimonas sp. AAP58	1523422	36.92	<0.01	0.21	0.23	1907
Aestuariiirgva litoralis	2650924	39.96	<0.01	0.48	0.48	1228
Aquabacterium pictum	2315236	35.99	<0.01	0.30	0.30	1934
Prochlorothrix hollandica PCC 9006 = CALU 1027	317619	28.44	<0.01	0.23	0.23	1137
actinobacterium SCGC AAA028-N15	938467	28.56	<0.01	0.45	0.45	128
Candidatus Nanopelagicus limnes	1884634	29.90	<0.01	0.55	0.87	120
Vulcaniibacterium thermophilum	1169913	30.88	<0.01	0.11	0.12	2038
Vulcaniibacterium gelatinicum	2598725	27.47	<0.01	0.12	0.13	1332
Pseudoxanthomonas	83618	27.27	<0.01	0.22	0.31	12
Methylocaldum sp. 14B	1912213	27.03	<0.01	0.22	0.20	421
Novosphingobium sediminis	707214	30.53	<0.01	0.31	0.31	1281
Novosphingobium kunmingense	1211806	26.37	<0.01	0.17	0.17	1451
Erythrobacter donghaensis	267135	26.55	<0.01	0.30	0.40	731
Erythrobacter sp. CCH5-A1	1768792	26.05	<0.01	0.49	0.58	85
Gemmobacter sp. HYN0069	2169400	24.52	<0.01	0.25	0.25	985
Rubrivivax benzoatilyticus JA2 = ATCC BAA-35	987059	25.21	<0.01	0.18	0.32	1597
Methylibium petroleiphilium PM1	420662	24.10	<0.01	0.18	0.16	789
Piscinibacter aquaticus	392597	26.77	<0.01	0.20	0.21	1654
Flaviumibacter sp. SB-02	2676868	24.10	<0.01	0.34	0.34	493
Candidatus Atelocyanobacterium thalassa	713887	15.88	<0.01	1.20	1.20	4
Anabaena cylindrica PCC 7122	272123	15.09	<0.01	0.19	0.21	451
Trichormus sp. NMC-1	1853259	18.16	<0.01	0.26	0.30	361
actinobacterium acIB-AMD-7	1504322	15.17	<0.01	0.32	0.35	61
Xanthomonas phaseoli pv. phaseoli	317013	18.29	<0.01	0.46	0.38	32
Perlucidibaca piscinae DSM 21586	1122951	18.15	<0.01	0.26	0.26	417
Novosphingobium sp. B 225	1961849	23.45	<0.01	0.18	0.18	992
Blastomonas sp. CCH8-A3	1768743	16.77	<0.01	0.66	1.07	11
Sphingomonas sp. IBVSS1	1985171	21.26	<0.01	0.31	0.40	309
Tabrizicola aquatica	909926	14.61	<0.01	0.18	0.21	517
Rhodobacter thermarum	2670345	22.08	<0.01	0.29	0.34	373
Rhodobacter flagellatus	2593021	15.39	<0.01	0.20	0.25	485

Gemmobacter caeruleus	2595004	15.61	<0.01	0.19	0.19	504
Rhodovulum visakhapatnamense	364297	18.35	<0.01	0.27	0.22	206
Rhodovulum strictum	58314	15.90	<0.01	0.34	0.32	819
Paracoccus sp. FO-3	1335059	18.38	<0.01	0.31	0.15	151
Aquincola tertiarycarbonis	391953	19.03	<0.01	0.11	0.11	1448
unclassified Methylibium	2633235	14.95	<0.01	0.16	0.18	408
Burkholderiales genera incertae sedis	224471	23.06	<0.01	0.15	0.15	83
Rivibacter subsaxonicus	457575	18.21	<0.01	0.13	0.13	1136
Schlegelella thermodepolymerans	215580	17.26	<0.01	0.12	0.12	1156
Limnohabitans sp. Hippo4	1826167	16.61	<0.01	0.19	0.25	310
Burkholderiales bacterium JOSHI_001	864051	17.15	<0.01	0.16	0.16	973
Vogesella mureinivorans	657276	16.81	<0.01	0.13	0.13	768
Gemmatimonas	173479	16.52	<0.01	12.55	14.04	82
Flavobacterium indicum GPTSA100-9 = DSM 17447	1094466	14.22	<0.01	0.12	0.12	306
Bacteroidetes bacterium SCGC AAA027-G08	938698	19.33	<0.01	0.39	0.39	88
'Nostoc azollae' 0708	551115	10.12	<0.01	0.15	0.15	228
actinobacterium SCGC AAA027-L06	913338	9.40	<0.01	0.19	0.28	37
actinobacterium acAcidi	1504320	5.79	<0.01	0.13	0.13	311
Actinobacteria bacterium IMCC26077	1848755	13.74	<0.01	0.22	0.22	177
Bacillus sp. JEM-1	1977090	8.70	<0.01	0.32	0.32	12
Aeromonas	642	12.30	<0.01	0.38	0.53	7
Novosphingobium subterraneum	48936	10.62	<0.01	0.15	0.11	241
Blastomonas natatoria	34015	9.76	<0.01	0.15	0.20	283
Sandarakinorhabdus sp. AAP62	1248916	8.74	<0.01	0.13	0.17	242
Sphingobium fluviale	2506423	8.43	<0.01	0.13	0.13	222
Erythrobacter dokdonensis DSW-74	1300349	10.28	<0.01	0.13	0.13	338
Erythrobacter sanguineus	198312	9.45	<0.01	0.13	0.13	281
Erythrobacter tepidarius	60454	12.42	<0.01	0.16	0.17	359
Erythrobacter cryptus DSM 12079	1122970	6.86	<0.01	0.11	0.11	162
alpha proteobacterium SCGC AAA027-C06	938624	5.69	<0.01	0.19	0.36	15
alpha proteobacterium SCGC AAA487-M09	938672	6.06	<0.01	0.22	0.23	13
alpha proteobacterium SCGC AAA027-J10	938631	4.84	<0.01	0.24	0.35	9
Tabrizicola piscis	2494374	12.20	<0.01	0.15	0.15	518
Rhodobacter veldkampii DSM 11550	1185920	11.73	<0.01	0.18	0.35	841
Rhodobacter blasticus	1075	9.68	<0.01	0.14	0.14	303
Gemmobacter aestuarii	1445661	11.13	<0.01	0.14	0.14	458
Gemmobacter aquatilis	933059	12.40	<0.01	0.16	0.16	395
Gemmobacter nectariphilus DSM 15620	1121271	11.93	<0.01	0.16	0.16	433
Fertoebacter nigrum	2656921	8.64	<0.01	0.10	0.10	369
Pseudorhodobacter sp. MZDSW-24AT	2052957	12.79	<0.01	0.17	0.17	404
Luteovulum ovatum	439529	7.77	<0.01	0.11	0.11	291
Rhabdaerophilum calidifontis	2604328	8.52	<0.01	0.13	0.13	266
Bradyrhizobium	374	5.78	<0.01	0.14	0.13	16
Rubrivivax gelatinosus IL144	983917	11.51	<0.01	0.10	0.25	517
Tepidimonas sediminis	2588941	11.11	<0.01	0.11	0.12	470
Inhella inkyongensis	392593	7.89	<0.01	0.10	0.10	318
Serpentinomonas raichei	1458425	10.80	<0.01	0.12	0.12	285
Comamonadaceae bacterium 2PF	2502197	8.69	<0.01	0.10	0.15	423
Comamonadaceae bacterium JGI 0001013-A16	1286843	6.91	<0.01	0.12	0.12	50
beta proteobacterium SCGC AAA027-I06	938781	7.19	<0.01	0.20	0.20	138
Thaueria sp. K11	2005884	9.07	<0.01	0.10	0.10	515
Methylophilus	16	5.30	<0.01	7.89	7.89	14
Crenobacter sp. GY 70310	2563443	12.02	<0.01	0.18	0.18	381
Chitinibacter sp. ZOR0017	1339254	8.03	<0.01	0.15	0.22	131
Vogesella urethralis	2592656	8.65	<0.01	0.14	0.15	255

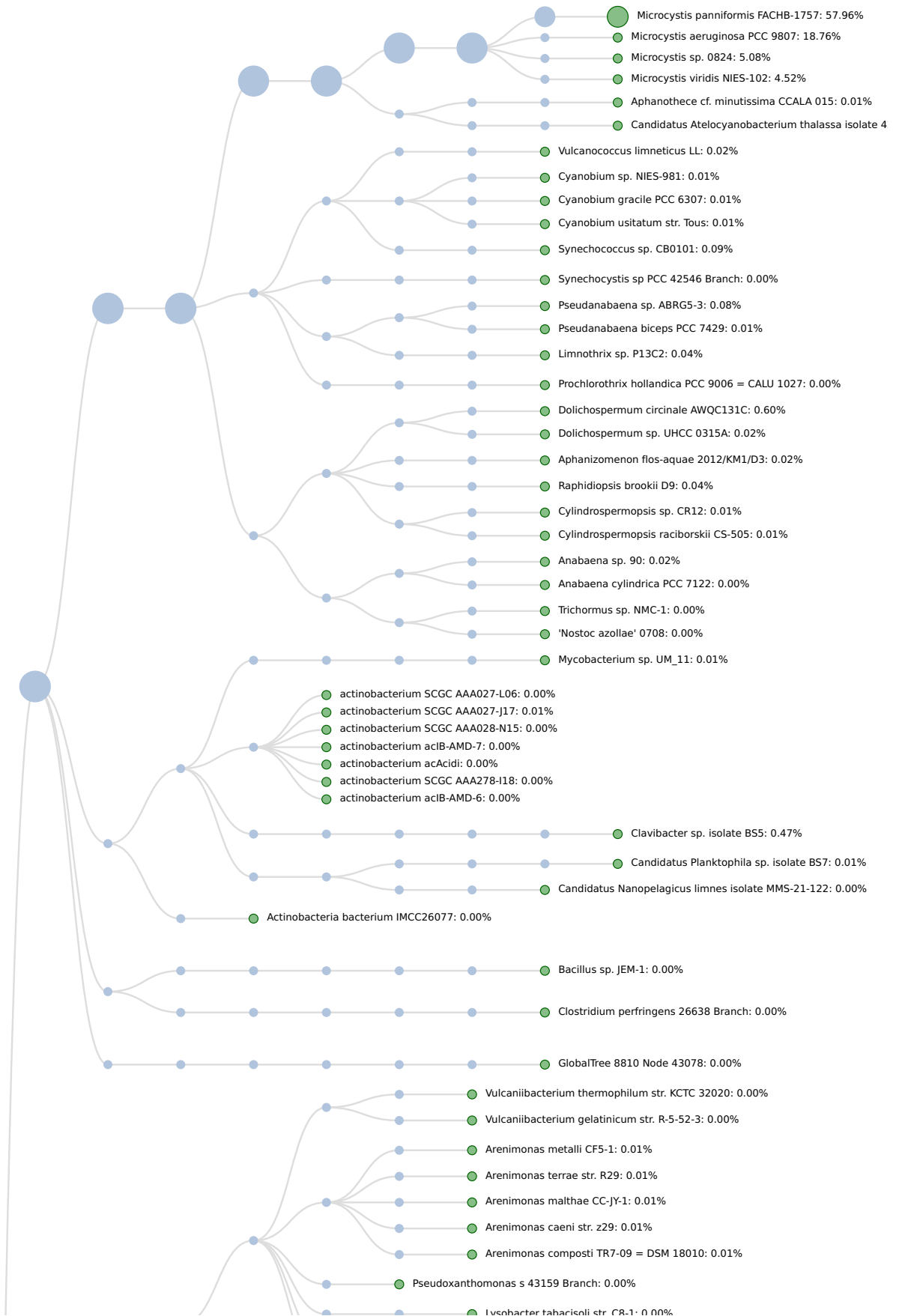
Opiritaceae bacterium TAV3	278958	10.55	<0.01	0.27	0.40	24
Opiritaceae bacterium TAV4	278959	5.53	<0.01	0.13	0.23	22
Flavobacterium tibetense	2233533	5.27	<0.01	0.11	0.12	70
Flavobacteria bacterium BAL38	391598	9.92	<0.01	0.10	0.24	137
Sediminibacterium salmonium NBRC 103935	1400522	10.57	<0.01	0.16	0.16	158
Candidatus Fervidibacteria	1383058	6.84	<0.01	0.75	0.75	2
unclassified Synechocystis	2640012	1.35	0.00	1.05	1.05	23
actinobacterium SCGC AAA278-I18	938557	4.71	0.00	0.15	0.15	18
actinobacterium acIB-AMD-6	1504321	1.38	0.00	0.11	0.11	16
Clostridium perfringens	1502	1.43	0.00	0.13	0.13	3
Sphaerobacteraceae	85002	2.21	0.00	6.90	6.90	6
alpha proteobacterium SCGC AAA027-L15	938633	1.09	0.00	0.12	0.21	5
alpha proteobacterium SCGC AAA028-C07	938639	2.73	0.00	0.11	0.11	10
alpha proteobacterium SCGC AAA028-D10	938641	1.49	0.00	0.11	0.19	7
Ignavibacteriales	795748	2.78	0.00	11.11	11.11	5

Bacteria

Tree Chart Taxonomy

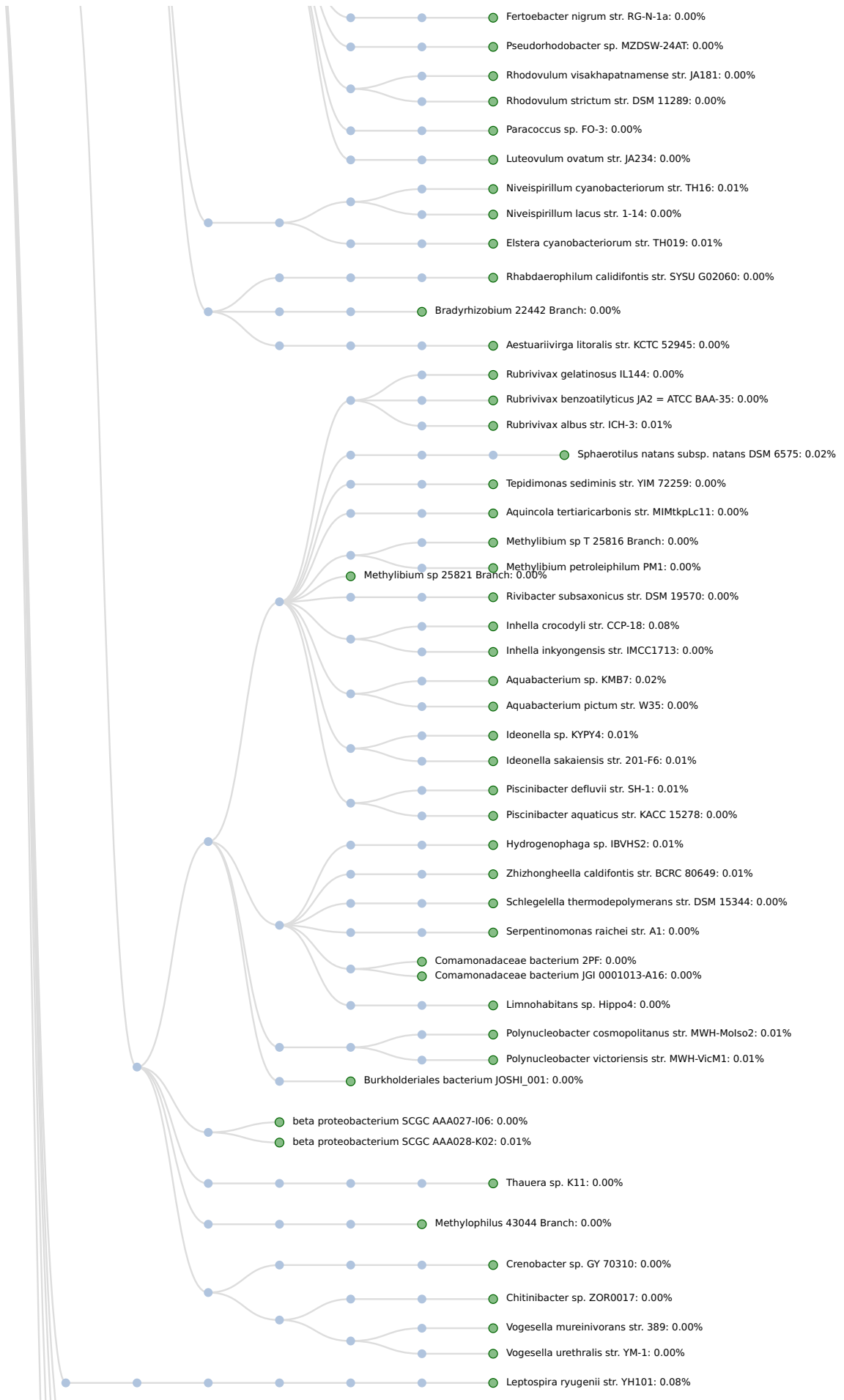
Sample\_2\_BWT2214

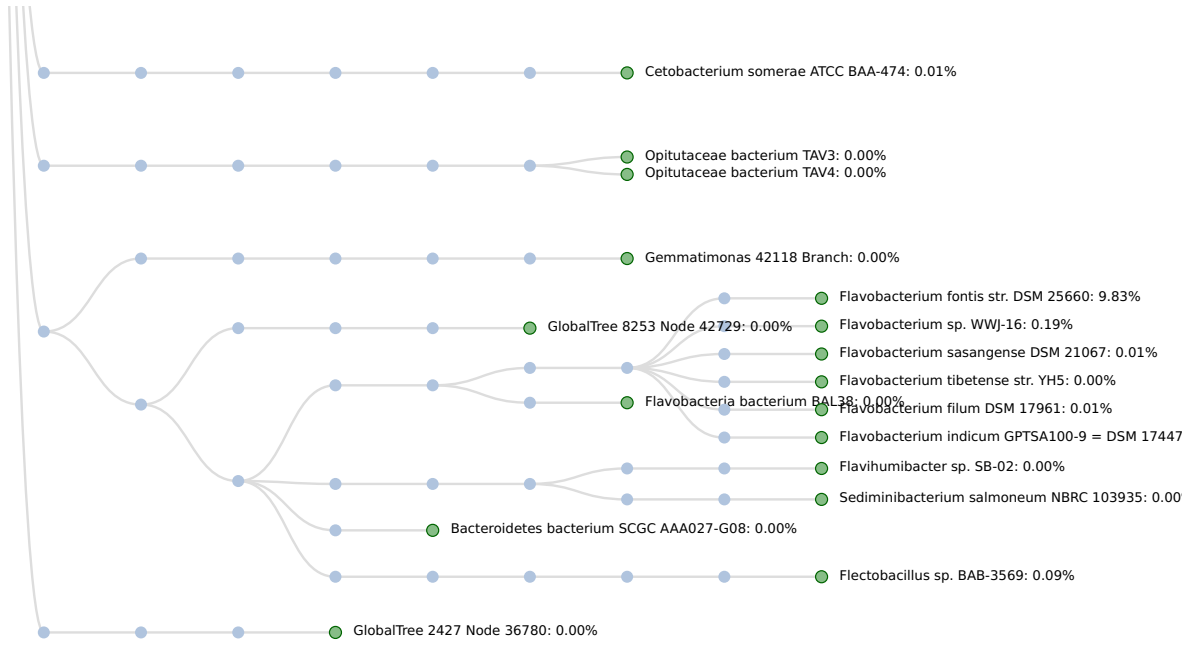
Total results









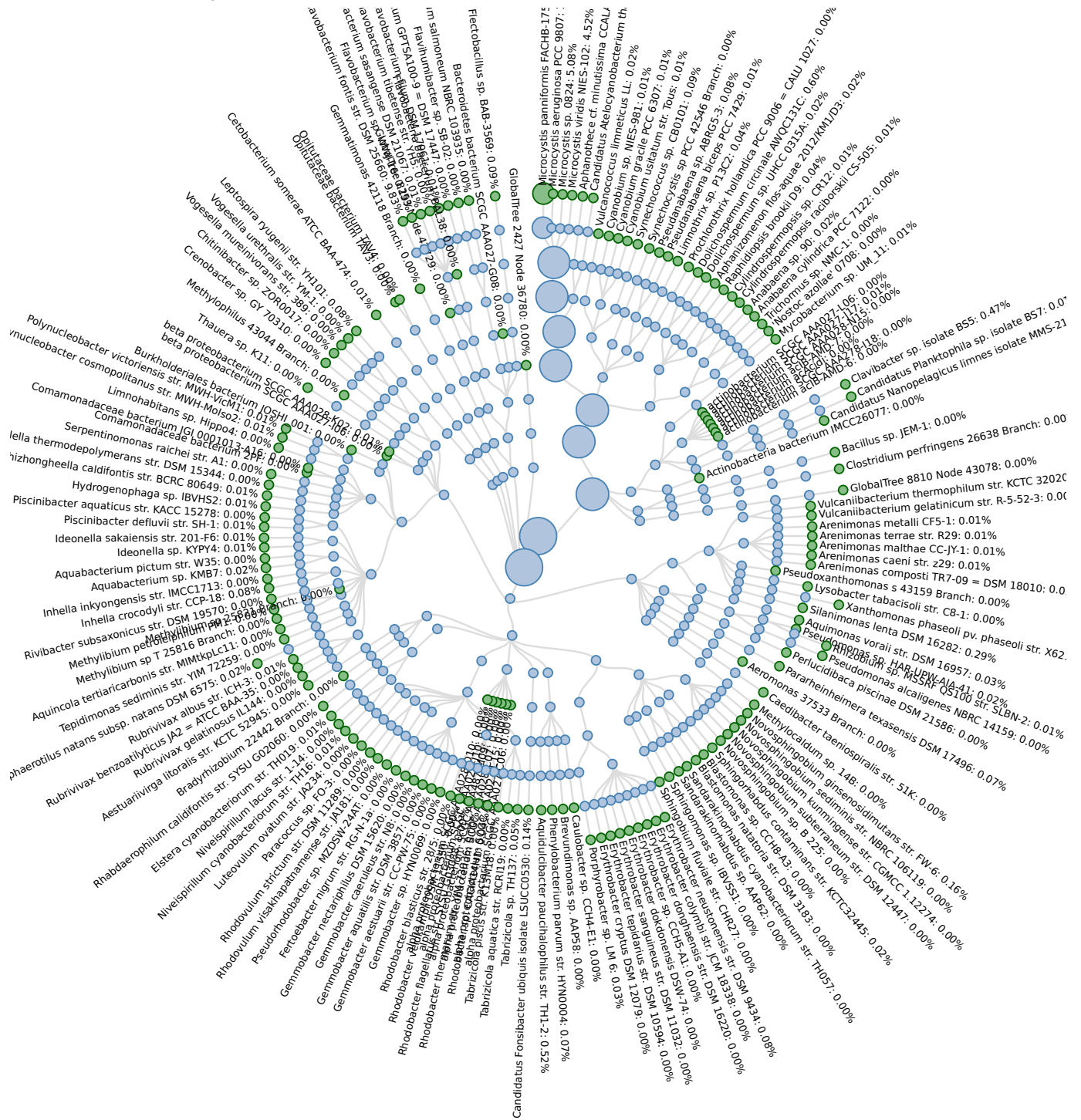


Bacteria

Radial Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results



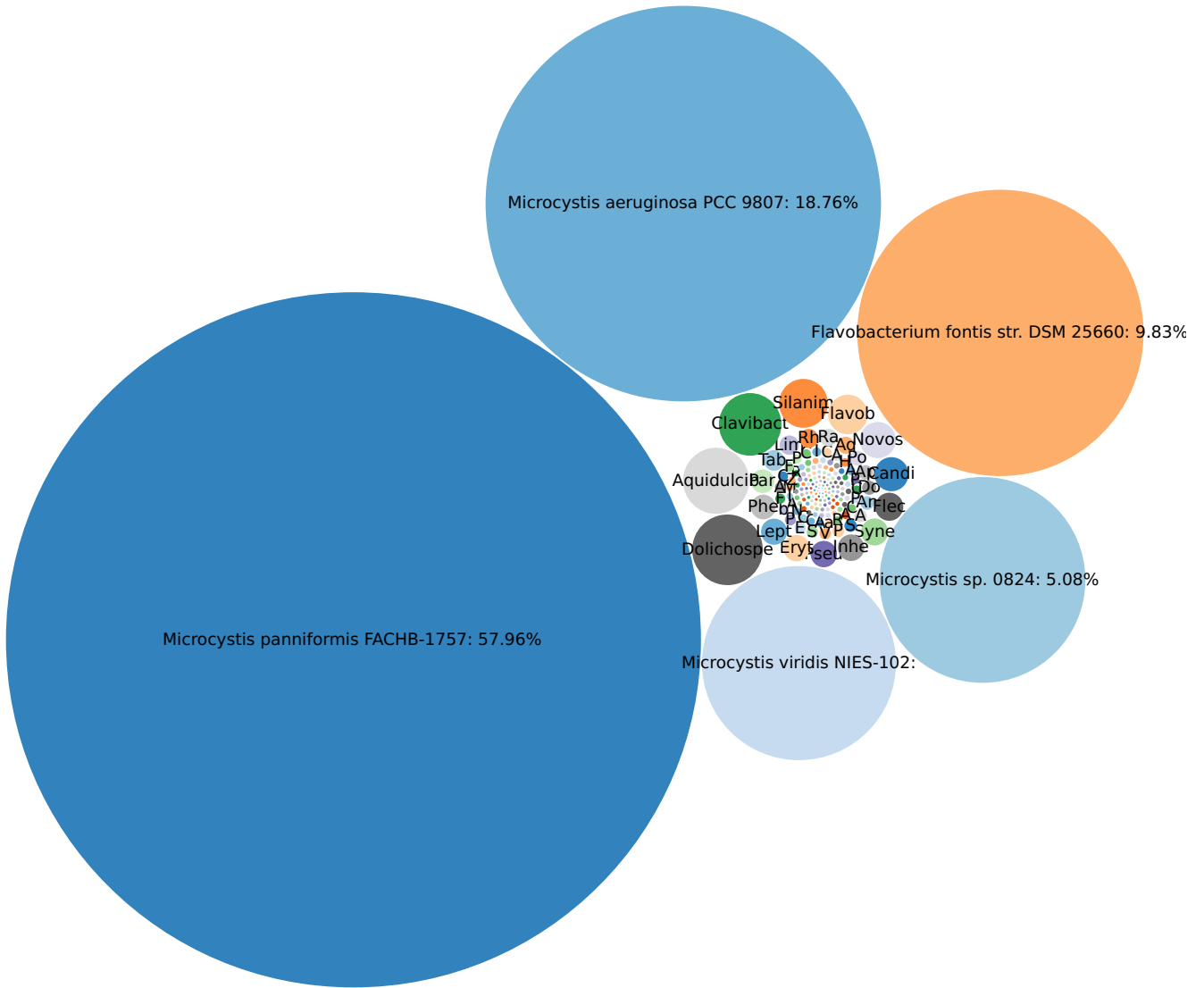


Bacteria

Bubble Chart Taxonomy

Sample\_2\_BWT2214

Total results



## Fungi

Sample\_2\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	37.179k

### Table

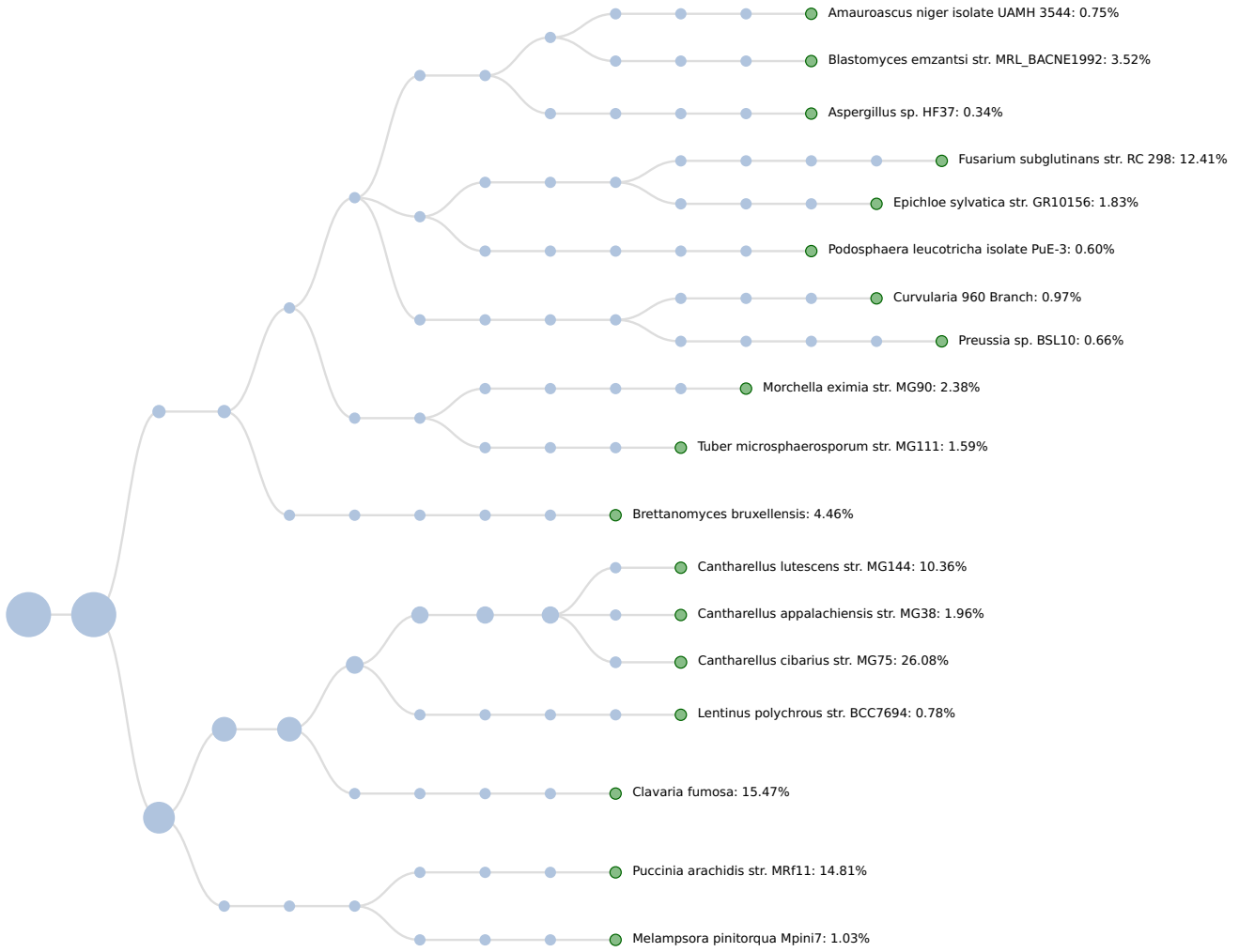
Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
<i>Cantharellus cibarius</i>	36066	24.46	26.08	0.08	0.10	2054
<i>Clavaria fumosa</i>	264083	14.51	15.47	0.10	0.30	1044
<i>Puccinia arachidis</i>	333523	13.89	14.81	0.07	0.13	1185
<i>Fusarium subglutinans</i>	42677	11.64	12.41	0.07	0.05	935
<i>Cantharellus lutescens</i>	104198	9.72	10.36	0.04	0.26	907
<i>Brettanomyces bruxellensis</i>	5007	4.18	4.46	0.02	0.03	434
<i>Blastomyces emzantsi</i>	2723674	3.30	3.52	0.03	0.04	273
<i>Morchella eximia</i>	1582338	2.23	2.38	0.03	0.01	198
<i>Cantharellus appalachiensis</i>	409893	1.84	1.96	0.02	0.21	187
<i>Epichloe sylvatica</i>	79593	1.72	1.83	0.03	0.02	1742
<i>Tuber microsphaerosporum</i>	1455713	1.49	1.59	0.02	0.02	155
<i>Melampsora pinitorqua Mpini7</i>	1298852	0.97	1.03	0.01	0.03	98
<i>Curvularia</i>	5502	0.91	0.97	0.01	0.01	27
<i>Lentinus polychrous</i>	292559	0.73	0.78	0.02	0.02	154
<i>Amauroascus niger</i>	89421	0.70	0.75	0.01	0.33	76
<i>Preussia sp. BSL10</i>	1712568	0.62	0.66	0.02	0.02	39
<i>Podosphaera leucotricha</i>	79249	0.56	0.60	0.01	0.01	55
<i>Aspergillus sp. HF37</i>	1960876	0.32	0.34	0.01	0.01	151

Fungi

Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results



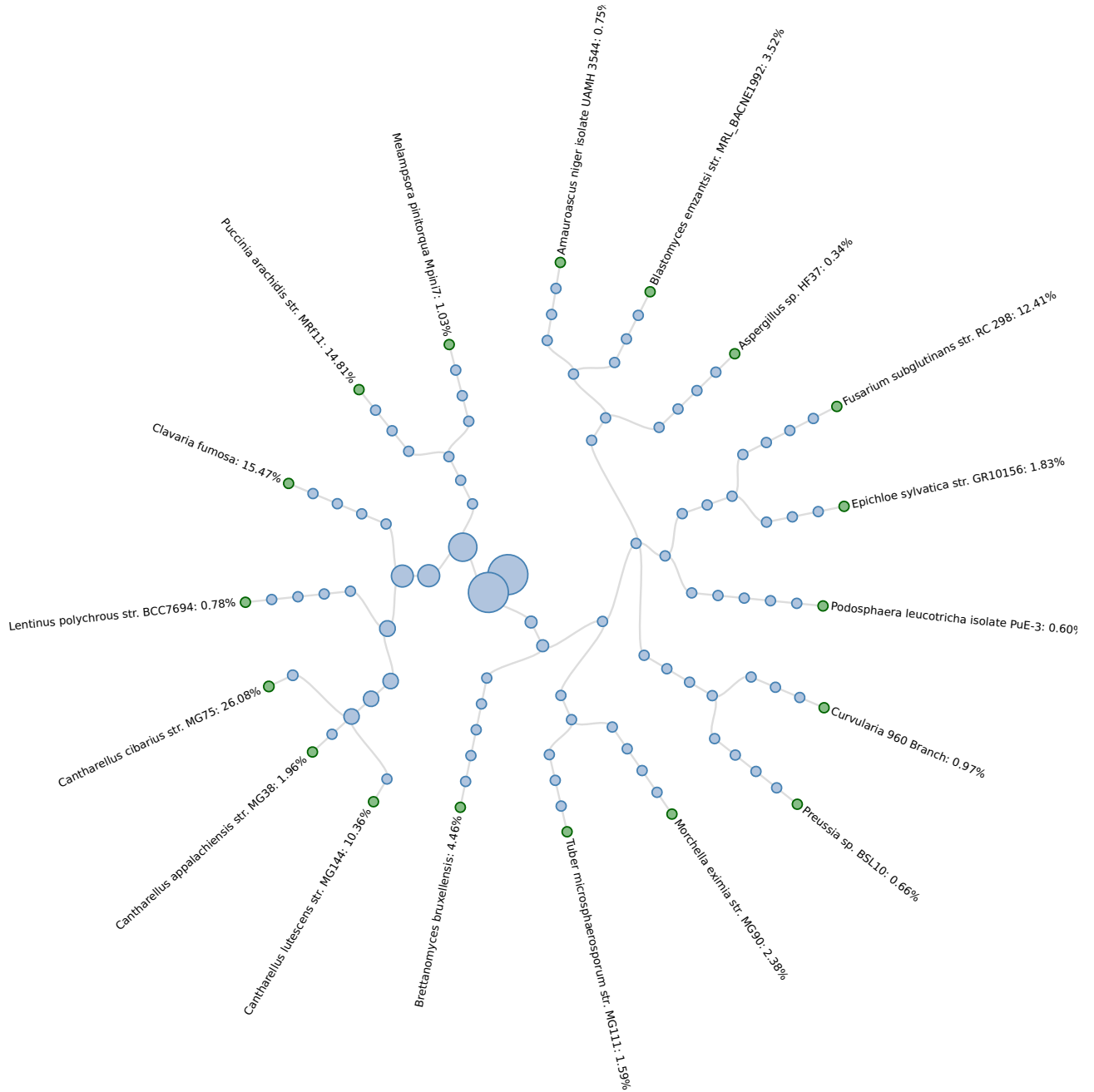


Fungi

Radial Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results

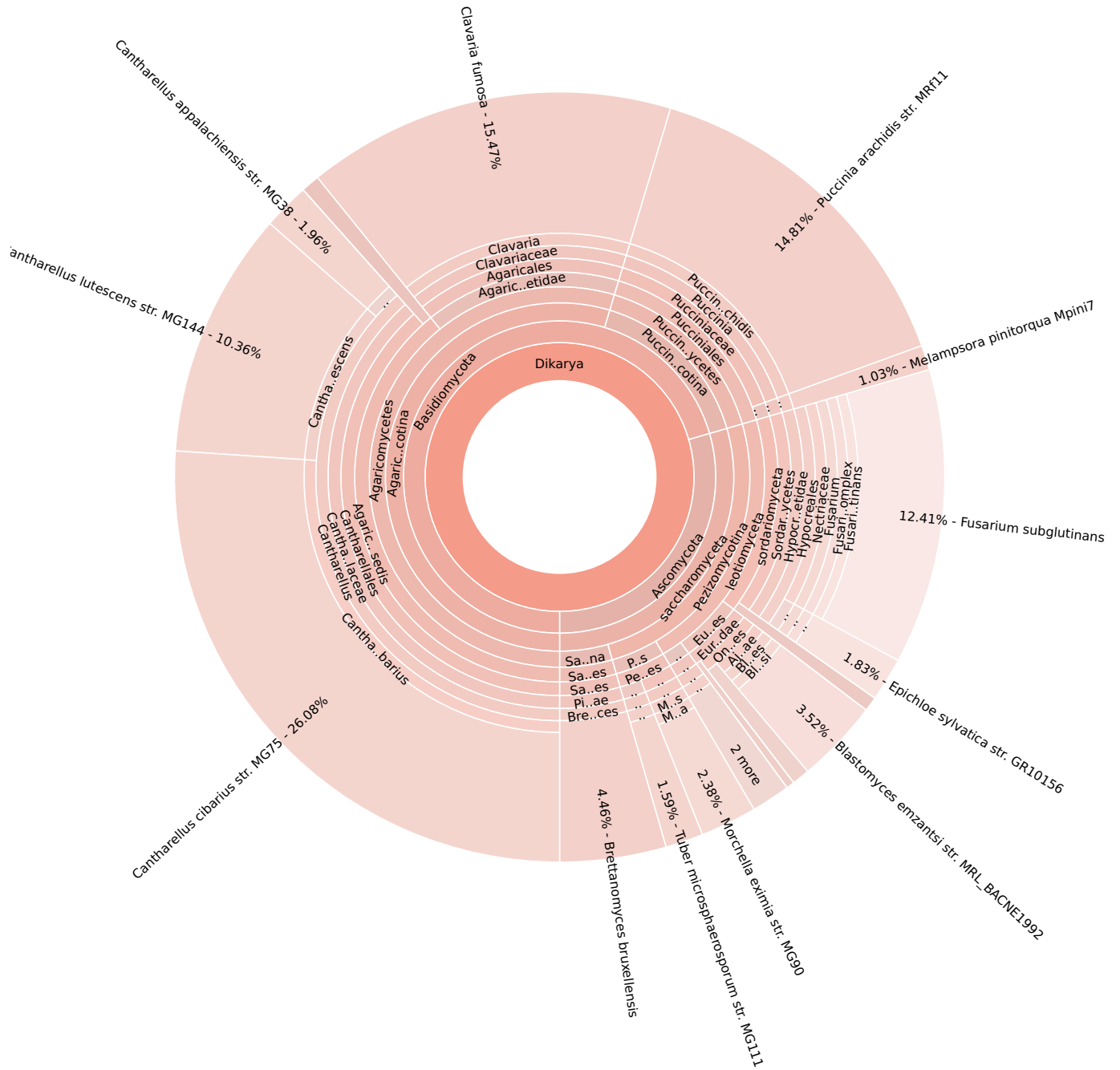


Fungi

Sunburst Chart Taxonomy

Sample\_2\_BWT2214

Total results

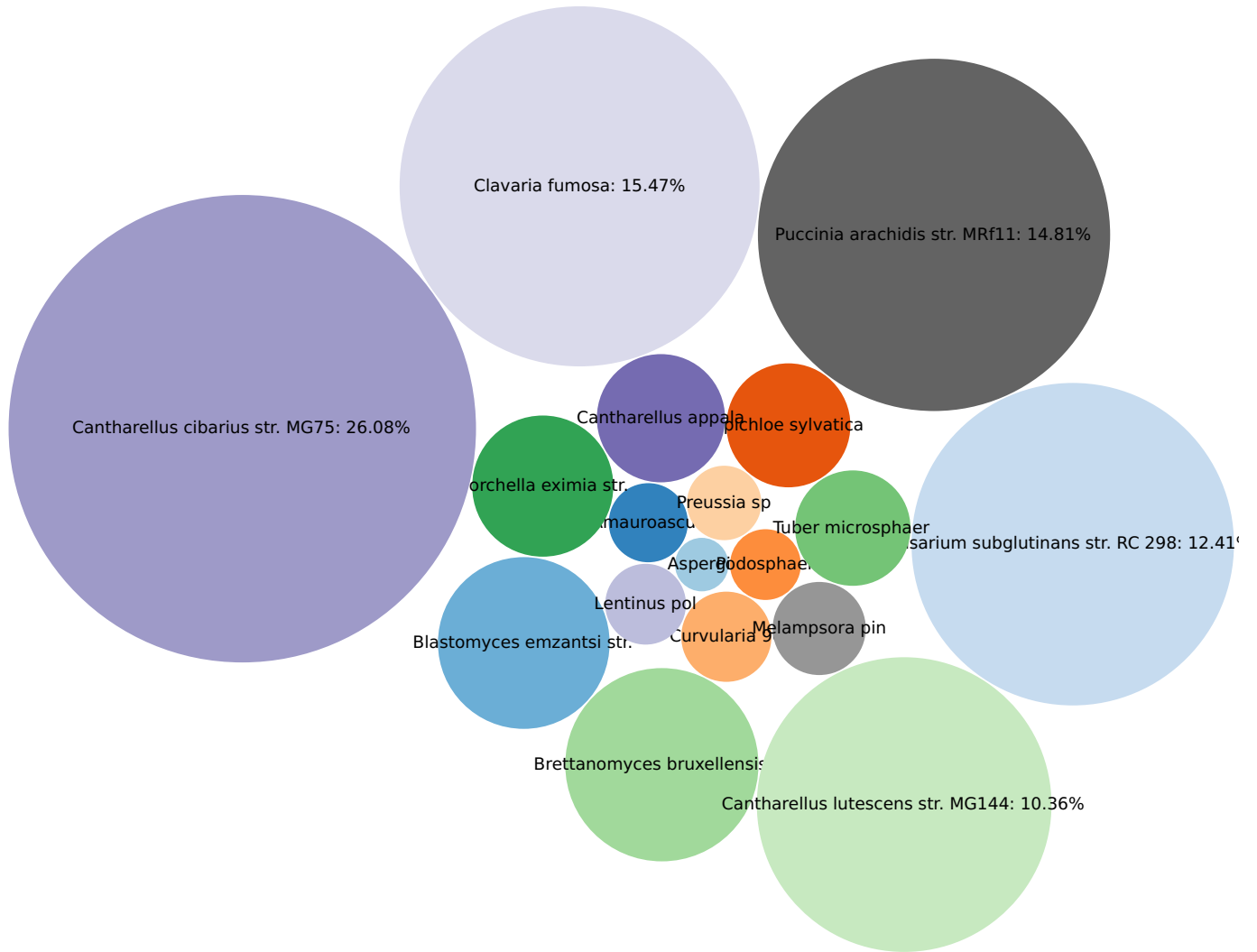


Fungi

Bubble Chart Taxonomy

Sample\_2\_BWT2214

Total results



## Protists

Sample\_2\_BWT2214

## Fasta/q details

Total results

Metric	Value
Hit	15.797k

## Table

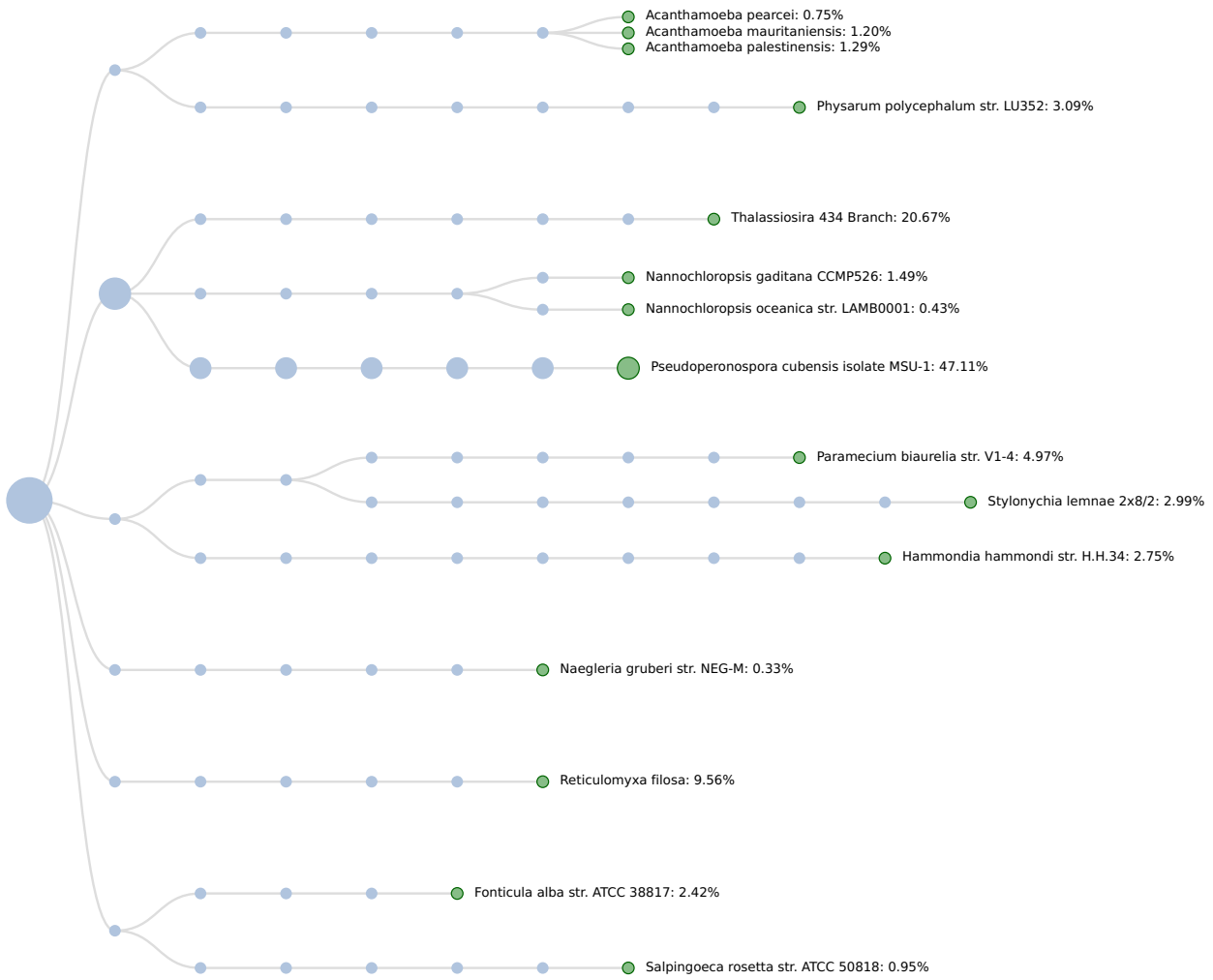
Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
<i>Pseudoperonospora cubensis</i>	143453	33.86	47.11	0.14	0.22	2575
<i>Thalassiosira</i>	35127	14.86	20.67	24.32	24.32	73
<i>Reticulomyxa filosa</i>	46433	6.87	9.56	0.06	0.09	522
<i>Paramecium biaurelia</i>	65126	3.57	4.97	0.03	0.03	319
<i>Physarum polycephalum</i>	5791	2.22	3.09	0.02	0.09	251
<i>Stylonychia lemnae 2x8/2</i>	755200	2.15	2.99	0.02	0.03	93
<i>Hammondia hammondi</i>	99158	1.98	2.76	0.03	0.04	169
<i>Fonticula alba</i>	691883	1.74	2.42	0.01	0.04	203
<i>Nannochloropsis gaditana CCMP526</i>	1093141	1.07	1.49	0.02	0.01	77
<i>Acanthamoeba palestinensis</i>	28015	0.93	1.29	0.02	0.09	85
<i>Acanthamoeba mauritaniensis</i>	196912	0.86	1.20	0.02	0.06	71
<i>Salpingoeca rosetta</i>	946362	0.68	0.95	0.02	0.23	40
<i>Acanthamoeba pearcei</i>	65662	0.54	0.75	0.02	0.03	8
<i>Nannochloropsis oceanica</i>	145522	0.31	0.43	0.01	0.07	278
<i>Naegleria gruberi</i>	5762	0.24	0.33	0.01	0.04	39

Protists

Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results

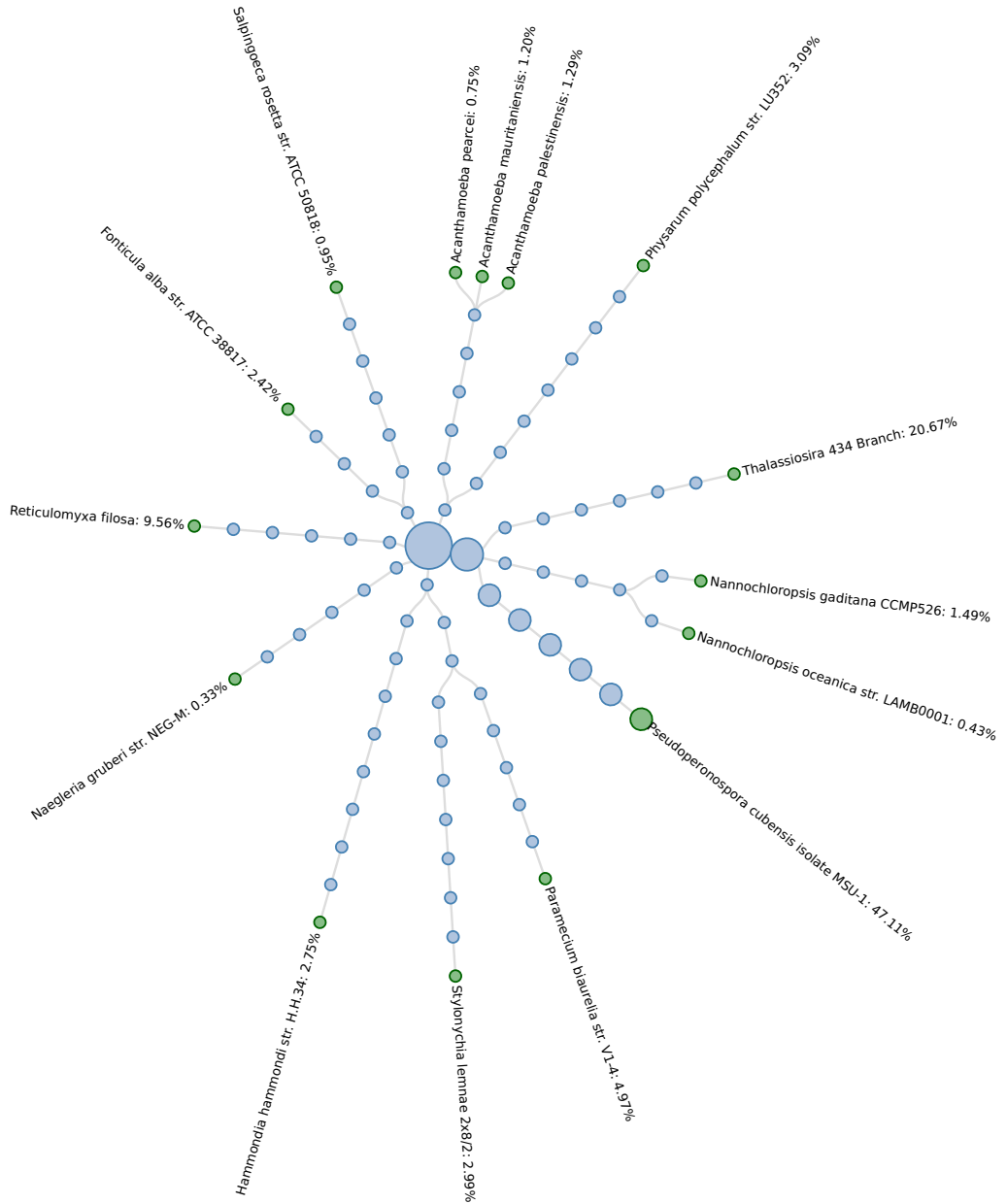


Protists

Radial Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results



Protists

Sunburst Chart Taxonomy

Sample\_2\_BWT2214

Total results

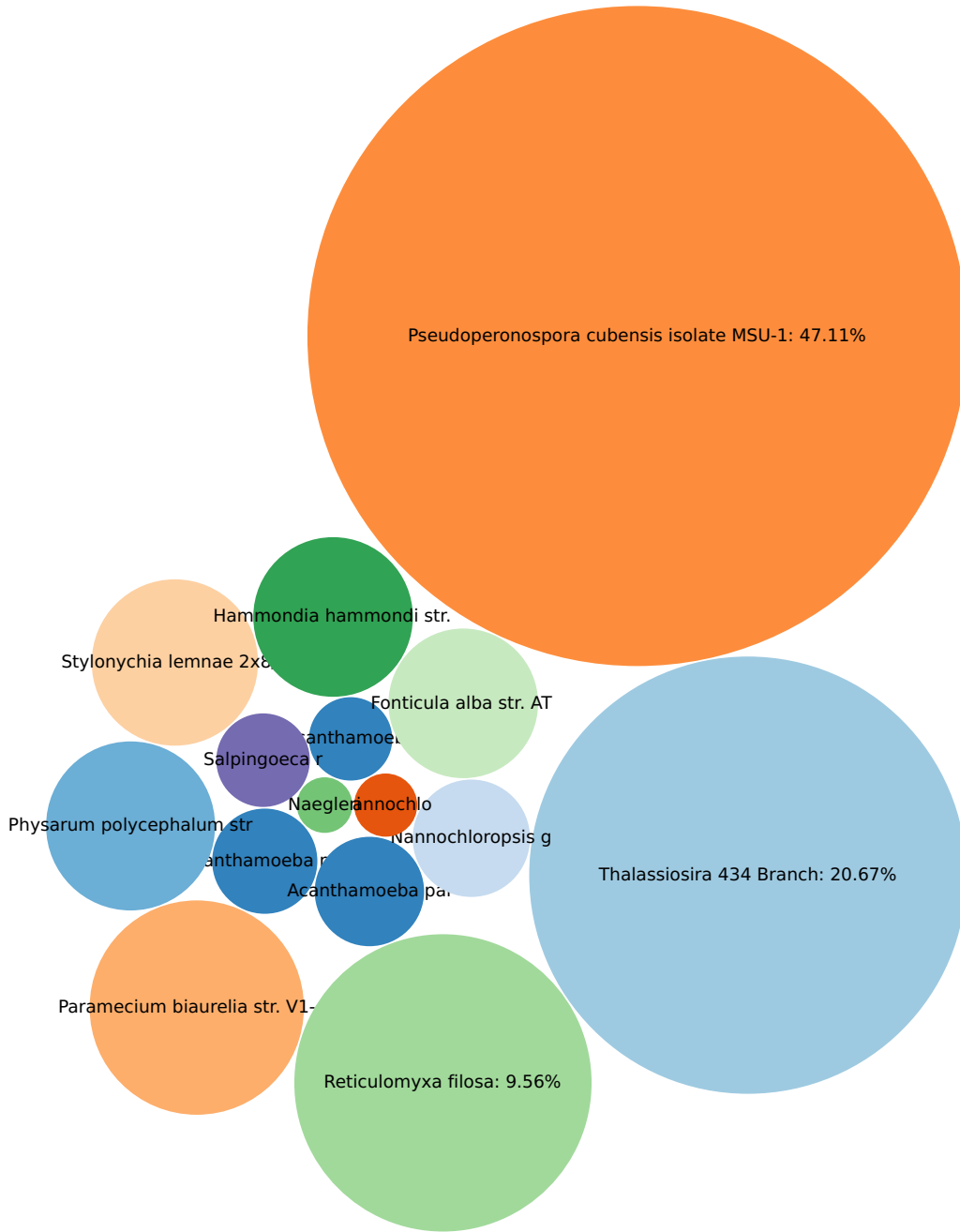


Protists

Bubble Chart Taxonomy

Sample\_2\_BWT2214

Total results





## Dark Matter (Beta)

Sample\_2\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	0.890M

Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Pseudanabaena sp.	1153	26206.62	41.61	45.52	45.75	449287
Burkholderiales bacterium UBA4657	1950779	5798.43	9.21	38.45	38.45	67918
Xanthomonadaceae bacterium	1926873	4887.30	7.76	4.78	4.78	135896
Burkholderiales bacterium UBA954	1950782	4425.09	7.03	33.32	33.32	32060
Novosphingobium sp. UBA6722	1946985	2873.99	4.56	17.42	17.42	43968
Pelagibacterales bacterium UBA4760	1951182	1779.14	2.83	19.98	19.98	2048
Chitinophagaceae bacterium	1869212	1709.48	2.71	19.17	24.63	3241
Proteobacteria bacterium UBA4656	1948178	1397.73	2.22	1.30	1.30	84487
Anabaena sp. UBA12330	2055756	1124.26	1.79	8.32	8.32	13249
Hyphomonadaceae bacterium	2026748	767.18	1.22	2.68	2.68	25481
Actinobacteria bacterium UBA3066	1948336	720.37	1.14	9.62	11.55	916
Chitinophagaceae bacterium UBA2467	1951550	699.69	1.11	7.03	9.85	1156
Burkholderiales bacterium UBA2470	1950772	594.64	0.94	5.56	8.77	1966
Chthonomonas sp. UBA2785	1946339	573.04	0.91	3.17	6.19	19
Actinobacteria bacterium	1883427	537.60	0.85	5.55	7.80	1216
Burkholderiales bacterium UBA2647	1950774	522.27	0.83	4.30	9.08	542
Porphyrobacter sp. UBA7686	1947080	522.07	0.83	4.05	4.05	9564
Actinobacteria bacterium UBA2236	1948325	521.64	0.83	7.48	11.11	543
Armatimonadetes bacterium	2033014	493.08	0.78	2.06	5.99	100
Arenimonas sp.	1872635	474.80	0.75	0.60	0.60	20648
Acidimicrobium sp.	1872112	462.72	0.73	5.06	6.33	1926
Lautropia sp.	2053568	425.45	0.68	5.17	9.39	605
Methylophilaceae bacterium	2030816	398.29	0.63	4.99	5.19	167
Flavobacterium sp. UBA6203	1946555	289.68	0.46	3.47	3.72	1020
Blastomonas sp. UBA7677	1946117	204.77	0.33	2.25	2.25	2659
unclassified Burkholderiales (miscellaneous)	80841	162.56	0.26	0.17	0.17	218
Chloroflexi bacterium	2026724	160.98	0.26	1.87	1.87	1361
Acidobacteria bacterium	1978231	144.69	0.23	2.06	2.06	492
Candidatus Microthrix	41949	129.44	0.21	1.15	1.15	35
Planctomycetaceae bacterium	2026779	125.97	0.20	1.37	1.37	3309
uncultured Collinsella sp.	165190	123.24	0.20	0.33	0.33	394
Verrucomicrobia bacterium UBA3063	1948252	122.99	0.19	1.24	1.36	1023
Pseudomonas sp. UBA6718	1947334	117.41	0.19	1.28	1.31	2259
unclassified Caulobacteraceae	81440	115.89	0.18	0.15	0.15	1136
unclassified Limnobacter	2630203	103.74	0.17	0.12	0.12	35
Proteobacteria bacterium UBA3065	1948161	101.58	0.16	0.95	0.50	692
unclassified Brevundimonas	2622653	95.47	0.15	0.23	0.23	232
Rheinheimera sp.	1869214	89.72	0.14	0.92	0.92	2325
Proteobacteria bacterium UBA964	1948225	78.09	0.12	0.73	0.73	2188
Methylocystaceae bacterium UBA4761	1952232	69.29	0.11	1.09	1.32	58
Alphaproteobacteria bacterium UBA2784	1948371	69.09	0.11	0.84	0.84	1237
Parachlamydiales bacterium	2052178	69.23	0.11	0.46	0.46	7831
Methylocystaceae bacterium UBA5192	1952233	68.28	0.11	1.06	1.27	119
Rhodanobacteraceae bacterium UBA7676	1953069	66.57	0.11	0.15	0.17	5182
Nitrosomonas sp. UBA6494	1946979	61.83	0.10	0.82	0.82	780
Gammaproteobacteria bacterium	1913989	60.74	0.10	0.23	0.23	2337
Sediminibacterium sp. UBA657	1947456	59.33	0.09	0.94	0.94	297

Aeromonas veronii	654	58.73	0.09	0.64	0.66	1327
Synechococcales bacterium UBA8647	2055797	56.78	0.09	0.79	0.42	159
Verrucomicrobia bacterium UBA2460	1948245	54.54	0.09	1.26	1.08	140
Polynucleobacter sp. UBA2464	1947076	53.02	0.08	0.63	0.35	286
Verrucomicrobia bacterium UBA2644	1948246	52.31	0.08	1.00	1.11	171
bacterium UBA951	1947788	50.18	0.08	0.85	0.85	150
Synechococcus sp. UBA8071	2055764	49.68	0.08	0.53	0.53	652
Ralstonia	48736	46.90	0.07	0.56	0.56	65
Phycisphaerales bacterium UBA4658	1951200	46.53	0.07	0.37	0.37	2420
unclassified Flavobacteriales (miscellaneous)	403978	46.03	0.07	3.03	3.03	2
Flammeovirgaceae bacterium UBA4659	1951784	42.58	0.07	0.39	0.39	1279
unclassified Polaromonas	2638319	41.54	0.07	0.30	0.30	54
unclassified Sphingopyxis	2614943	41.15	0.07	1.26	1.26	11
Rhodobacteraceae bacterium UBA6273	1952813	40.11	0.06	0.39	0.39	1478
Rhodanobacteraceae bacterium UBA703	1953068	39.20	0.06	0.14	0.16	499
Curvibacter sp.	1888168	37.37	0.06	0.34	0.35	1469
Synechococcales bacterium UBA12195	2055798	35.72	0.06	0.58	0.36	80
Comamonadaceae bacterium	1871071	36.14	0.06	0.26	0.26	1615
Bacteroidetes bacterium UBA955	1953175	35.13	0.06	0.52	0.52	367
Stenotrophomonas sp.	69392	33.70	0.05	0.12	0.13	1637
unclassified Candidatus Microthrix	2642024	33.10	0.05	0.70	0.68	22
uncultured Lachnobacterium sp.	1339173	33.28	0.05	0.16	0.16	414
Flavobacteriales bacterium UBA2426	1950978	33.25	0.05	0.38	0.38	1054
Candidatus Accumulibacter	327159	33.61	0.05	0.18	0.18	5
Cyanobacteria bacterium UBA6965	1947892	32.77	0.05	0.63	0.41	78
Xylella sp. UBA6001	1947786	32.99	0.05	0.12	0.13	2335
Brevundimonas sp. UBA7534	1946138	31.37	0.05	0.13	0.14	1809
Pseudomonas aeruginosa group	136841	30.75	0.05	0.51	0.51	24
Achromobacter	222	31.00	0.05	0.13	0.14	231
Synechococcales bacterium UBA10510	2055795	30.26	0.05	1.53	0.82	223
Ramlibacter tataouinensis	94132	30.11	0.05	0.15	0.15	1642
bacterium UBA5577	1947808	30.20	0.05	0.35	0.35	478
Cyanobacteria bacterium UBA7373	1947895	29.84	0.05	0.33	0.33	514
Pseudomonas stutzeri	316	29.35	0.05	0.50	0.53	107
Rhodobacteraceae bacterium UBA1943	1952798	29.54	0.05	0.29	0.29	1023
Rhodobacteraceae bacterium UBA6141	1952809	29.45	0.05	0.32	0.32	861
unclassified Bradyrhizobium	2631580	27.90	0.04	0.25	0.25	208
Spartobacteria bacterium UBA5019	1948749	27.93	0.04	0.39	0.40	512
Planctomycetaceae bacterium UBA4655	1952734	26.78	0.04	0.33	0.33	966
Tepidimonas sp. UBA997	1947650	25.90	0.04	0.22	0.22	881
Ignavibacteria bacterium UBA961	1948688	24.88	0.04	0.45	0.45	139
Delftia acidovorans	80866	25.23	0.04	0.17	0.15	746
Rhodocyclaceae bacterium UBA2250	1952837	24.97	0.04	0.19	0.19	1089
Rhodobacter sp.	1062	24.90	0.04	0.25	0.25	922
Alicyclophilus sp. UBA7619	1946005	24.29	0.04	0.22	0.24	1340
Aquabacterium sp. UBA666	1946043	23.19	0.04	0.28	0.39	465
Comamonadaceae bacterium UBA2334	1951601	23.39	0.04	0.24	0.24	1029
Gemmobacter sp.	1898957	22.71	0.04	0.29	0.20	187
Crocinitomicaceae bacterium	2026728	21.11	0.03	0.34	0.34	215
Burkholderiales bacterium UBA4200	1950777	21.11	0.03	0.38	0.21	136
Comamonadaceae bacterium UBA6202	1951611	21.28	0.03	0.20	0.20	890
Prochlorococcus sp. UBA3999	1947246	20.49	0.03	0.34	0.27	84
Comamonadaceae bacterium UBA4962	1951608	20.77	0.03	0.18	0.20	1199
Acidimicrobiaceae bacterium UBA6895	1951341	19.95	0.03	0.23	0.13	191
Planctomycetes bacterium UBA2386	1948127	20.38	0.03	0.19	0.19	1120

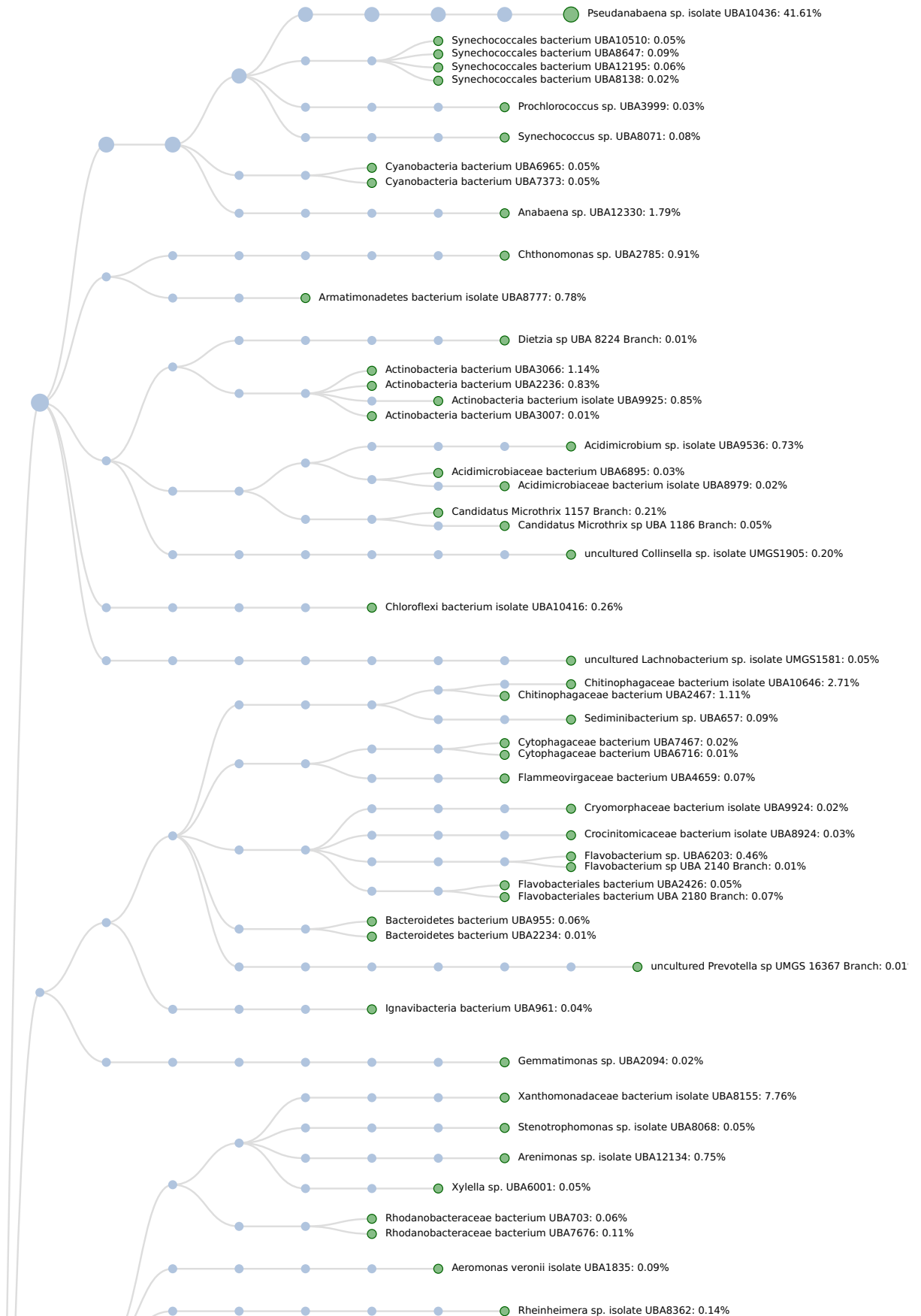
Hylemonella sp. UBA6679	1946611	16.39	0.03	0.20	0.20	483
unclassified Achromobacter	2626865	16.17	0.03	0.11	0.16	81
Gemmatimonas sp. UBA2094	1946567	15.59	0.03	0.22	0.22	508
Caulobacter vibrioides	155892	15.97	0.03	0.11	0.11	1169
Proteobacteria bacterium UBA2646	1948154	15.90	0.03	0.26	0.33	70
Burkholderiales bacterium UBA1834	1950768	15.29	0.02	0.20	0.19	234
Acidovorax sp.	1872122	14.94	0.02	0.15	0.15	846
Rhizobiaceae bacterium UBA3490	1953065	15.09	0.02	0.20	0.20	576
Citricella sp.	1960284	15.36	0.02	0.11	0.11	288
Rhodobacteraceae bacterium UBA996	1952817	15.12	0.02	0.17	0.17	668
Cryomorpaceae bacterium	1898111	14.68	0.02	0.23	0.23	239
Comamonas	283	14.78	0.02	0.13	0.13	133
Sphingomonas sp. UBA1000	1947531	14.47	0.02	0.13	0.13	516
Caulobacteraceae bacterium UBA6225	1951540	14.25	0.02	0.11	0.16	570
Rhodobacteraceae bacterium UBA6236	1952812	13.62	0.02	0.18	0.18	323
Paracoccus sp. UBA3880	1947052	12.92	0.02	0.14	0.14	420
Cytophagaceae bacterium UBA7467	1951636	12.23	0.02	0.32	0.44	33
Rhodobacteraceae bacterium	1904441	12.25	0.02	0.11	0.11	548
Rhodobacteraceae bacterium UBA6796	1952814	11.71	0.02	0.14	0.15	409
Opitutae bacterium UBA953	1948744	12.24	0.02	0.18	0.18	333
Synechococcales bacterium UBA8138	2055796	11.48	0.02	0.13	0.13	168
Comamonas aquatica	225991	10.51	0.02	0.13	0.14	392
Acidimicrobiaceae bacterium	2024894	10.08	0.02	0.13	0.13	496
Thiomonas sp. UBA7699	1947694	10.32	0.02	0.11	0.11	438
Hydrogenophaga sp.	1904254	10.23	0.02	0.27	0.25	205
Erythrobacter sp. UBA1916	1946469	9.78	0.02	0.11	0.11	370
Roseovarius sp. UBA6217	1947396	10.19	0.02	0.12	0.13	357
unclassified Rhodocyclaceae	75788	9.43	0.01	0.31	0.31	1614
Citromicrobium sp. UBA1476	1946344	9.31	0.01	0.11	0.12	321
Proteobacteria bacterium UBA2411	1948150	9.52	0.01	0.17	0.20	198
Bdellovibrionaceae bacterium UBA2651	1961509	9.74	0.01	0.33	0.23	16
Bdellovibrionaceae bacterium UBA2466	1961508	9.23	0.01	0.29	0.23	20
Verrucomicrobia bacterium UBA3062	1948251	9.48	0.01	0.16	0.16	149
Verrucomicrobiales	48461	9.18	0.01	0.12	0.12	127
Cytophagaceae bacterium UBA6716	1951634	9.09	0.01	0.39	0.46	9
Rhodiferax sp. UBA6134	1947380	8.76	0.01	0.12	0.12	231
unclassified Dietzia	2617939	8.04	0.01	0.37	0.37	6
Acidovorax delafieldii	47920	8.16	0.01	0.11	0.11	376
Methylocystaceae bacterium UBA2095	1952231	7.48	0.01	0.17	0.81	50
Geobacter sp. UBA4074	1961393	7.00	0.01	0.18	0.19	629
Verrucomicrobiaceae bacterium UBA2429	1952942	7.06	0.01	0.16	0.12	48
Phycisphaerales bacterium UBA2396	1951192	6.65	0.01	0.10	0.10	285
Actinobacteria bacterium UBA3007	1948330	6.50	0.01	0.17	0.17	31
Bacteroidetes bacterium UBA2234	1953144	6.57	0.01	0.13	0.13	99
uncultured Prevotella sp.	159272	6.18	0.01	0.14	0.14	10
unclassified Flavobacterium	196869	5.27	<0.01	0.32	0.32	4
unclassified Pseudomonas	2583993	4.82	<0.01	0.19	0.19	3
Legionellales bacterium	2026754	3.91	<0.01	0.12	0.12	24
Proteobacteria bacterium UBA2462	1948151	3.61	<0.01	0.13	0.31	13
Pseudomonas kunmingensis	1211807	3.10	<0.01	0.12	0.10	11
Erythrobacteraceae bacterium UBA5049	1951760	2.43	<0.01	0.28	0.21	14

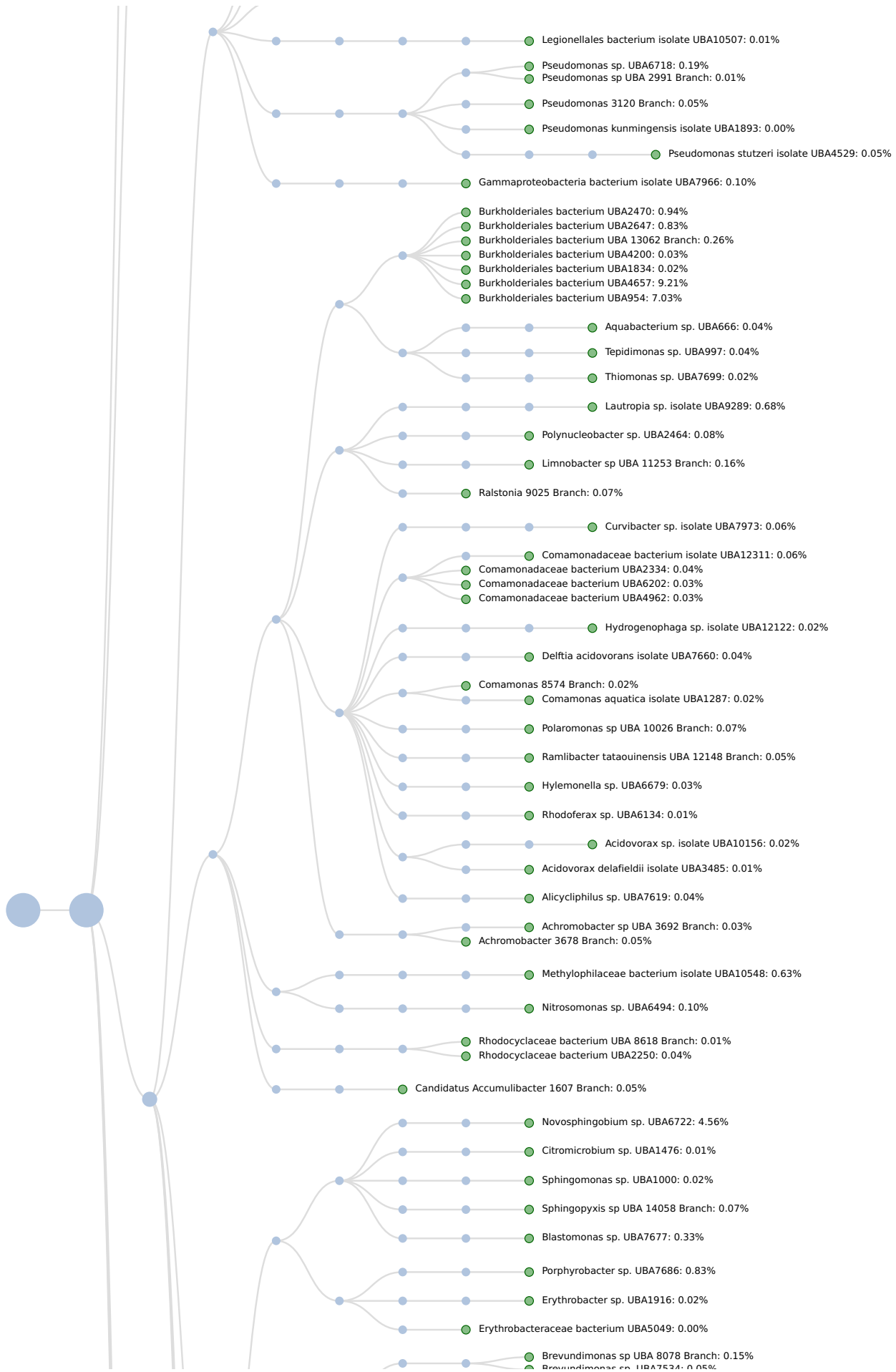
Dark Matter (Beta)

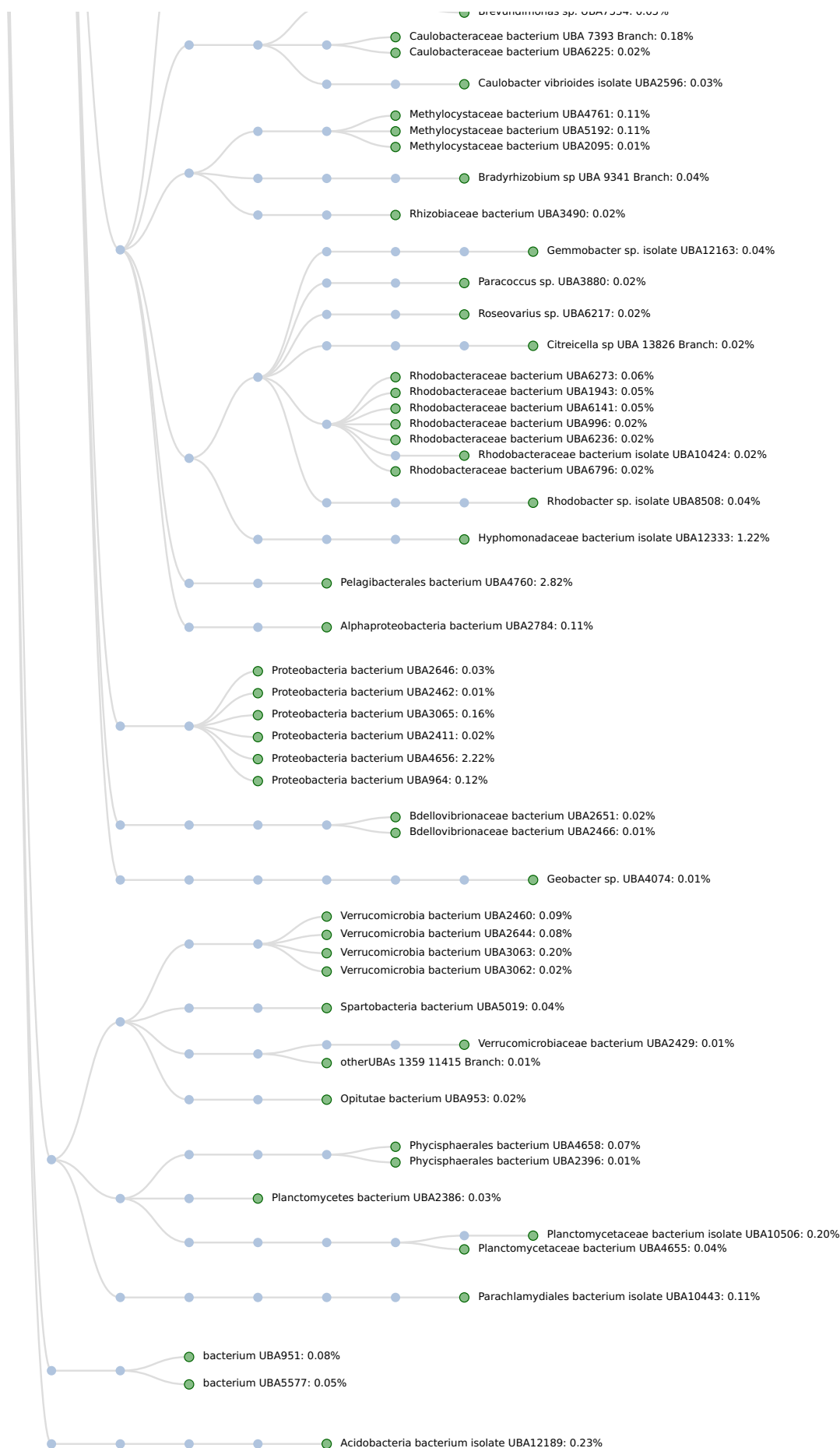
Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results







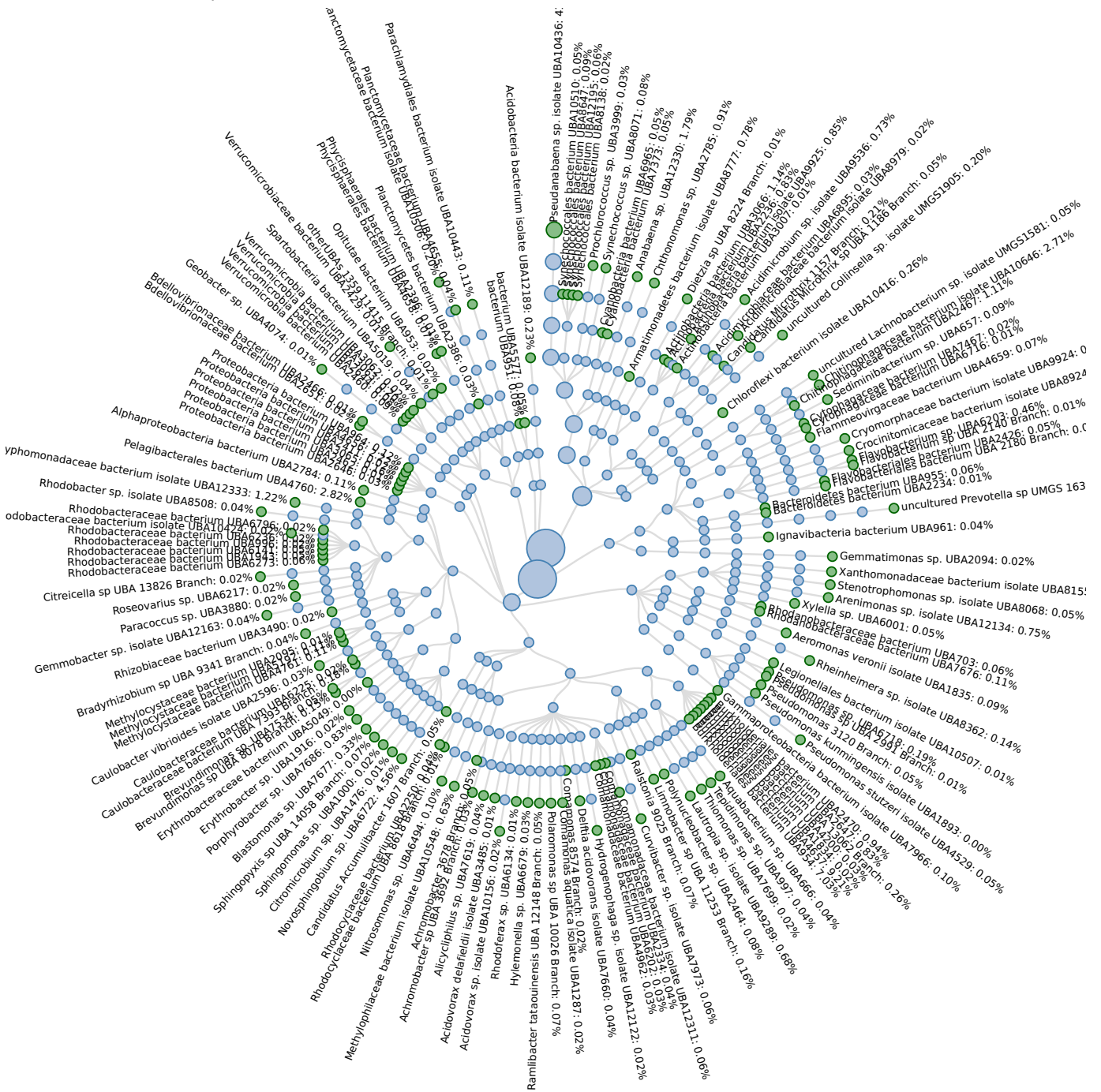


# Dark Matter (Beta)

Radial Tree Chart Taxonomy

# Sample\_2\_BWT2214

Total results





Dark Matter (Beta)

Sunburst Chart Taxonomy

Sample\_2\_BWT2214

Total results





## Phages

Sample\_2\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	0.299M

### Table

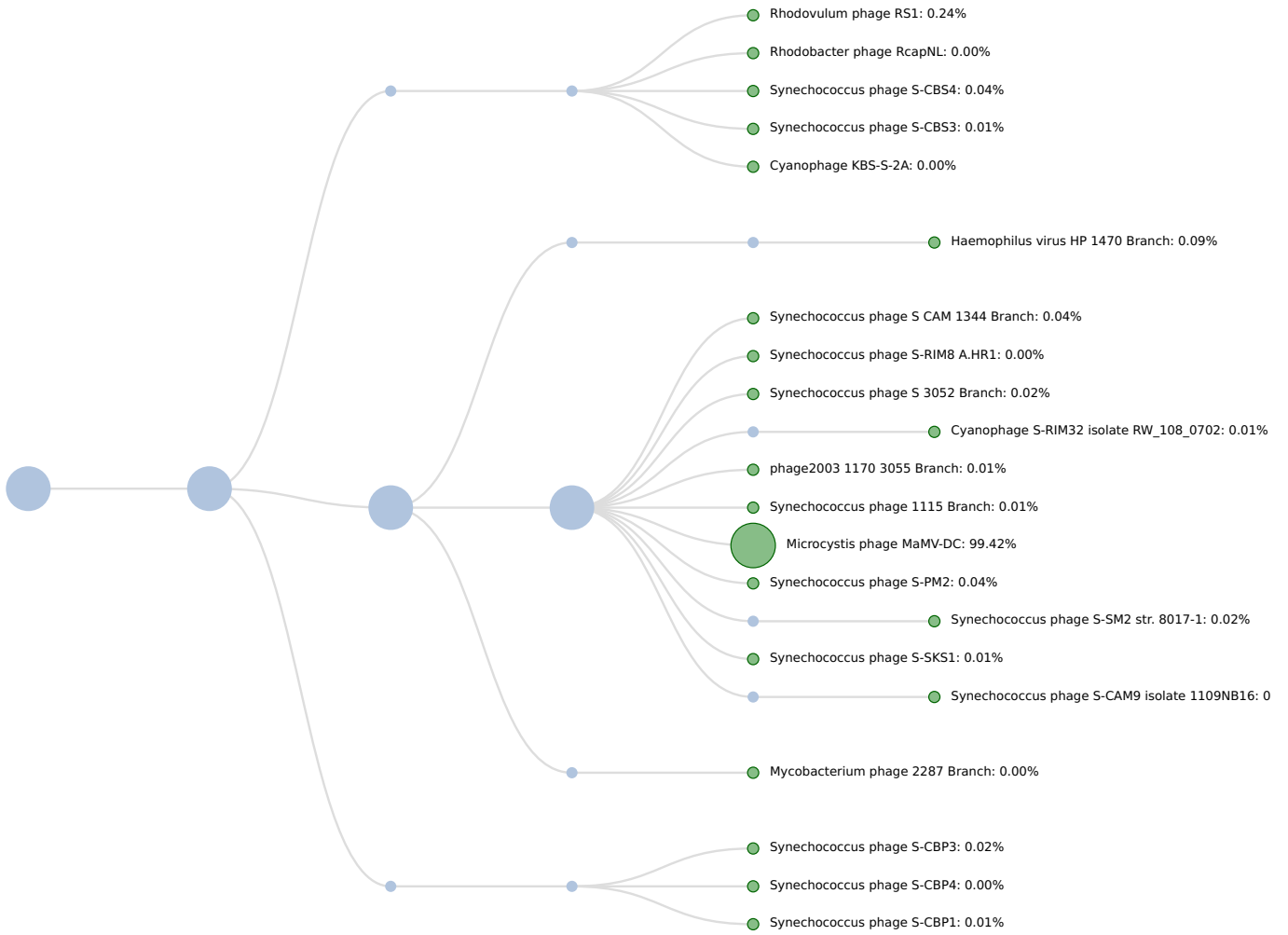
Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis phage MaMV-DC	1357715	168650.24	99.42	44.34	44.34	3874534
Rhodovulum phage RS1	754056	408.60	0.24	1.92	1.92	146
Hpunavirus	1196844	154.39	0.09	0.25	0.25	929
Synechococcus phage S-PM2	238854	74.82	0.04	0.58	0.60	2094
Synechococcus phage S-CBS4	756275	67.87	0.04	0.63	0.63	924
unclassified Myoviridae	196896	63.41	0.04	0.55	0.55	102
unclassified Myoviridae	196896	32.03	0.02	0.37	0.37	140
Synechococcus phage S-SM2	444860	30.33	0.02	0.33	0.37	774
Synechococcus phage S-CBP3	756276	25.54	0.01	0.33	0.61	255
Synechococcus phage S-SKS1	754042	22.76	0.01	0.27	0.32	578
unclassified Myoviridae	196896	16.90	0.01	0.16	0.16	47
Synechococcus phage S-CBP1	1273711	16.81	0.01	0.30	0.52	5169
Cyanophage S-RIM32	1278479	14.85	<0.01	0.12	0.19	582
Synechococcus phage S-CBS3	753085	12.87	<0.01	0.21	0.21	97
Synechococcus phage S-CAM9	1883369	12.67	<0.01	0.16	0.21	365
unclassified Myoviridae	196896	10.36	<0.01	0.19	0.19	33
Cyanophage KBS-S-2A	889953	5.13	<0.01	0.14	0.14	31
Synechococcus phage S-RIM8 A.HR1	869724	4.97	<0.01	0.10	0.21	487
Synechococcus phage S-CBP4	754059	5.87	<0.01	0.13	0.37	52
Rhodobacter phage RcapNL	1131316	2.27	<0.01	0.11	0.11	106
Bixzunavirus	680114	0.42	0.00	0.38	0.38	12

Phages

Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results

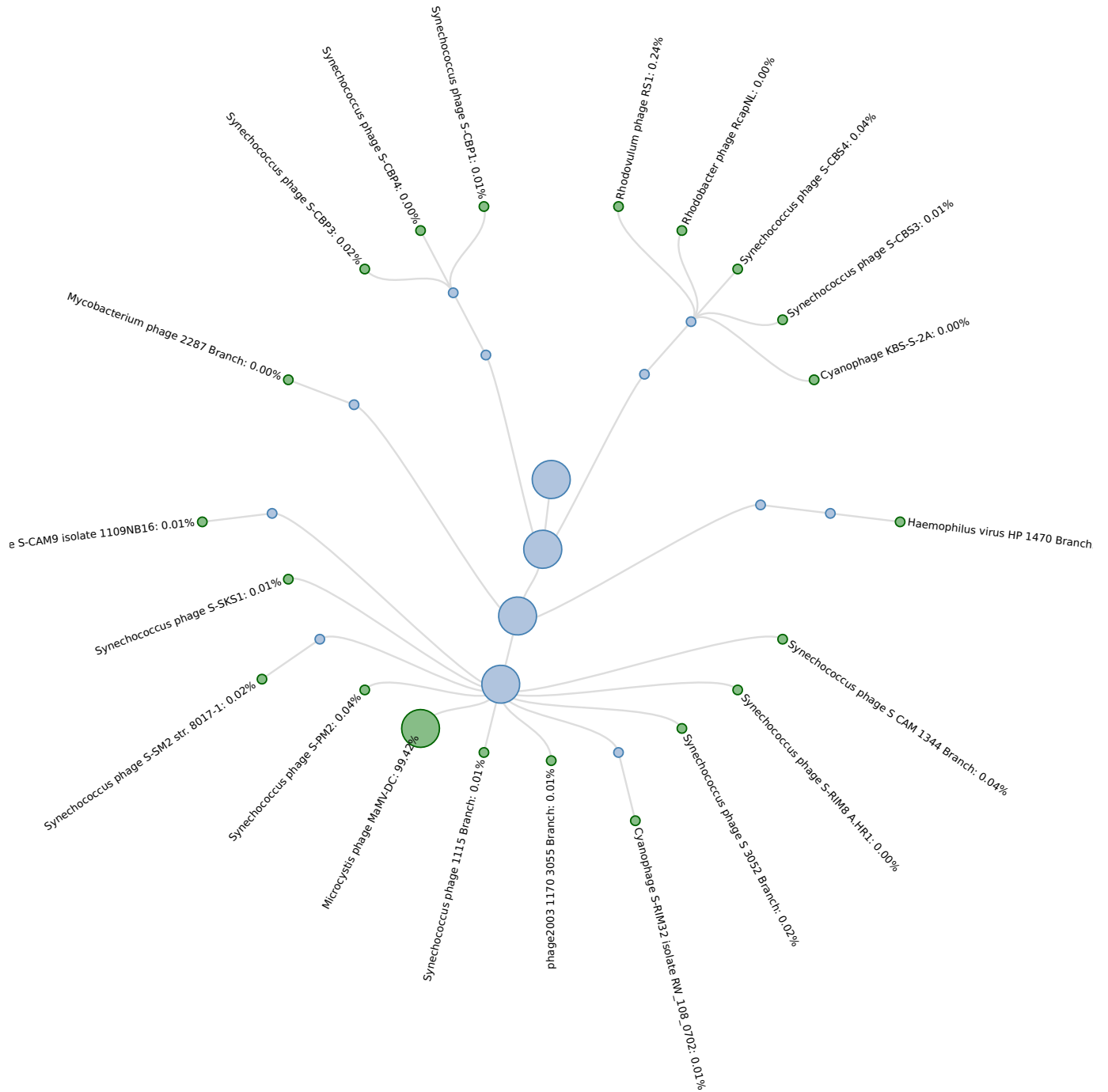


# Phages

Radial Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results

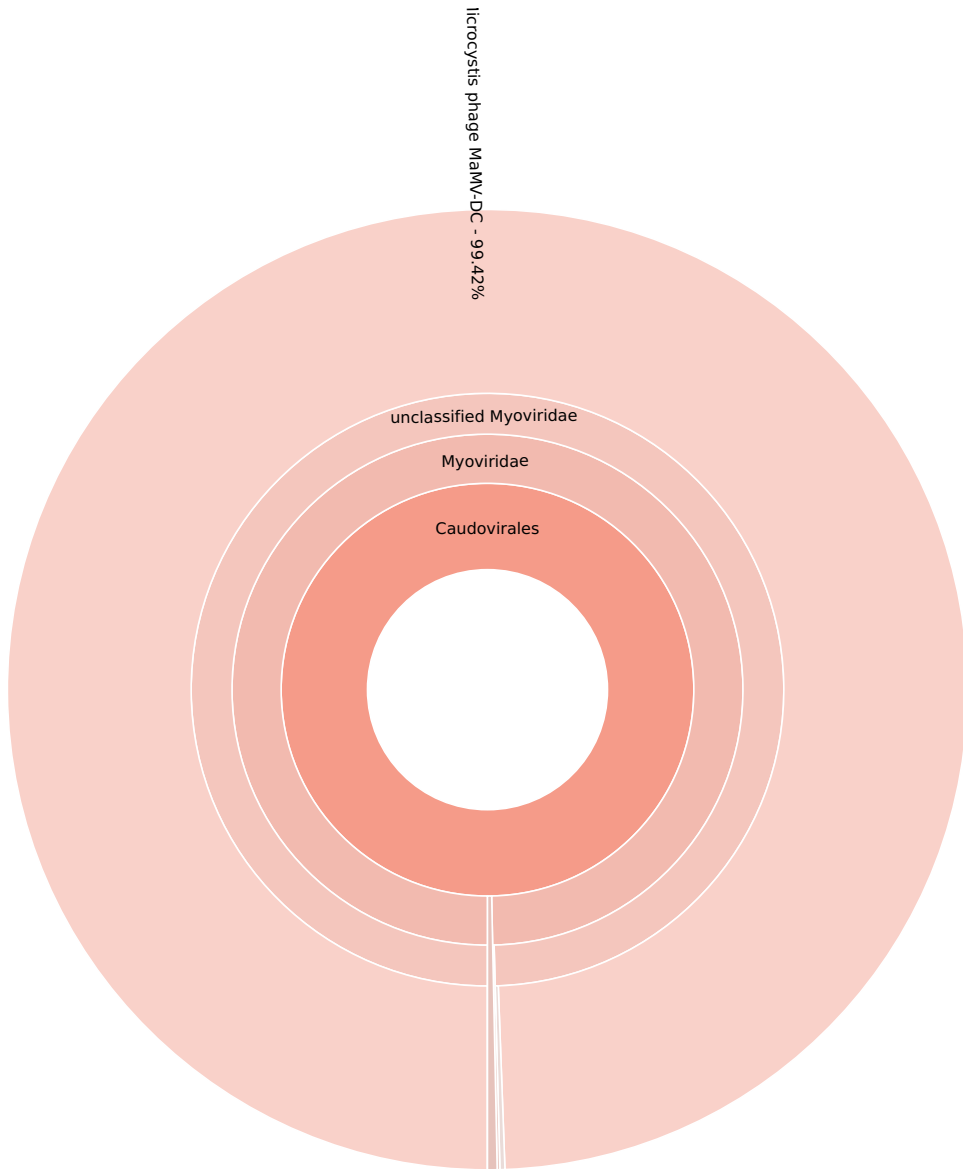


# Phages

Sunburst Chart Taxonomy

Sample\_2\_BWT2214

Total results

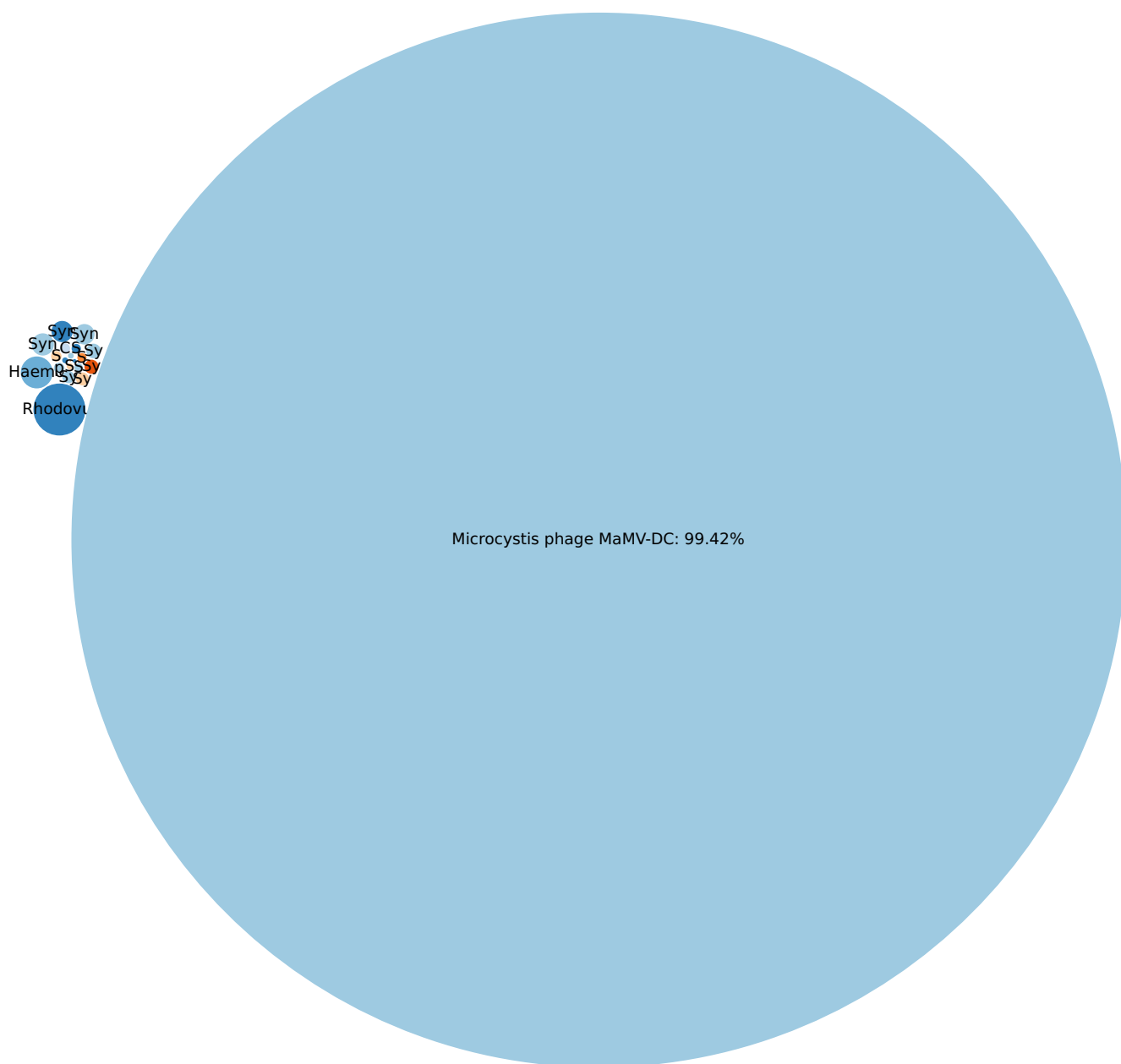


# Phages

Bubble Chart Taxonomy

Sample\_2\_BWT2214

Total results



## Viruses

Sample\_2\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	0.302M

### Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Microcystis virus Ma-LMM01	340435	189253.57	99.99	49.10	49.10	4330601
Rosellinia necatrix partitivirus 2	859651	11.86	<0.01	0.35	0.35	19

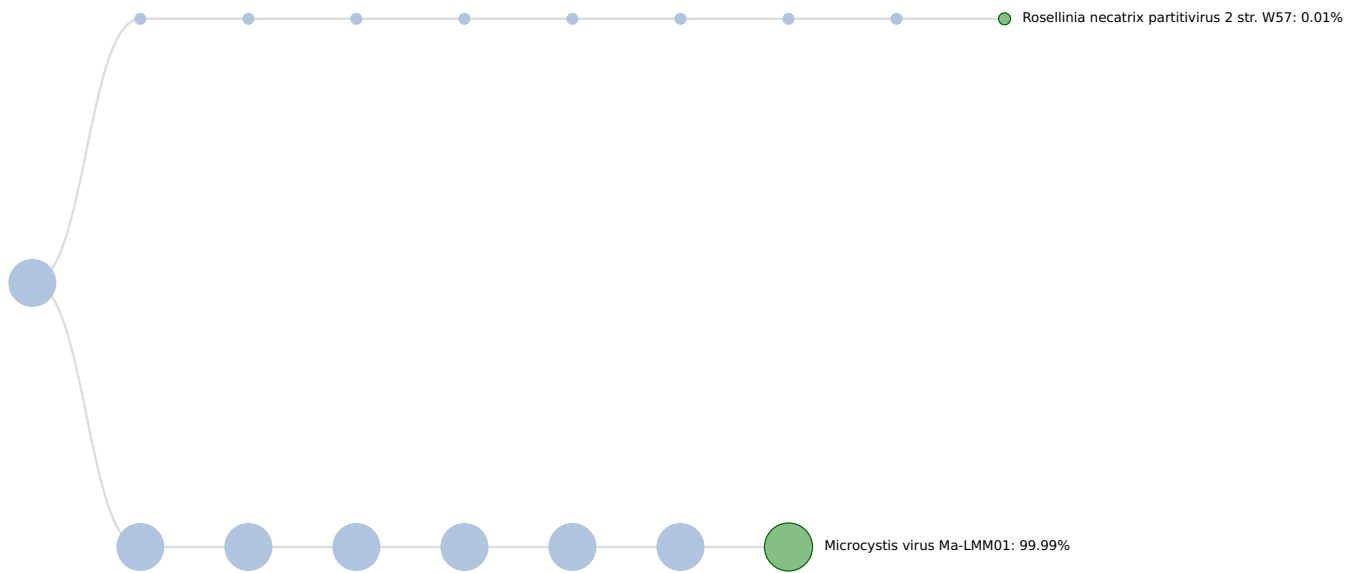


Viruses

Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results



# Viruses

Radial Tree Chart Taxonomy

Sample\_2\_BWT2214

Total results

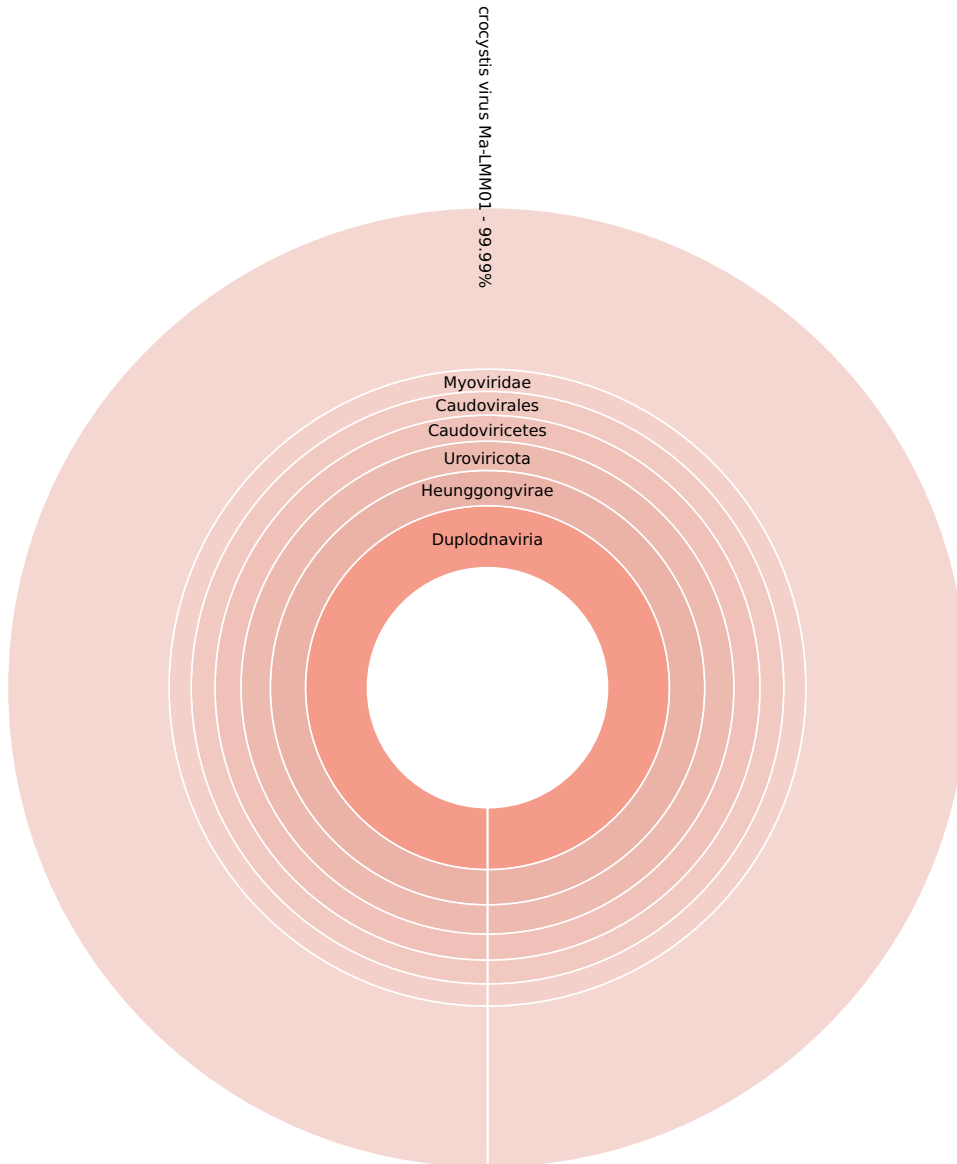


# Viruses

Sunburst Chart Taxonomy

Sample\_2\_BWT2214

Total results

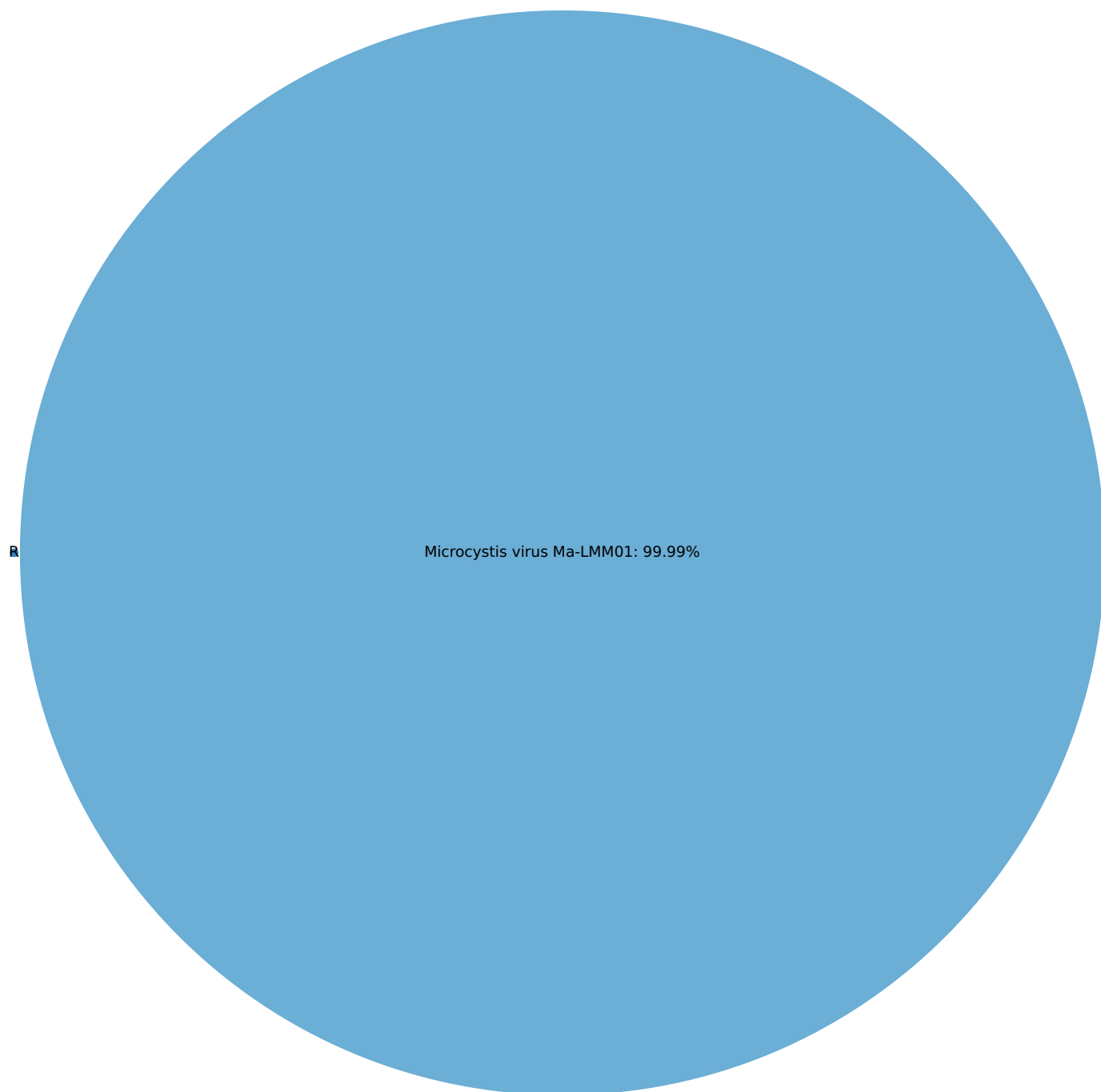


Viruses

Bubble Chart Taxonomy

Sample\_2\_BWT2214

Total results



## Virulence Factors

Sample\_2\_BWT2214

Fasta/q details

Total results

Metric	Value
Hit	1.950k

### Table

Name	Tax ID	Abundance Score	Relative Abundance %	Unique Matches %	Total Matches %	Frequency
Pseudomonas aeruginosa GENE int1	-	10642.65	40.92	100.00	100.00	342
Enterobacter aerogenes GENE tniC	-	7396.85	28.44	80.56	80.56	592
Proteus mirabilis GENE sul1	-	2445.54	9.40	62.12	62.12	41
Enterobacter aerogenes GENE traj	-	2167.88	8.34	33.01	33.01	130
Klebsiella pneumoniae GENE tnpA	-	1152.48	4.43	23.70	23.70	85
Enterobacter aerogenes GI 1572570	-	992.34	3.82	14.09	14.09	190
Enterobacter aerogenes GENE tniB	-	615.39	2.37	10.45	10.45	82
Enterobacter aerogenes GENE tniA	-	593.56	2.28	12.69	12.69	79

## Disclaimer

CosmosID is designed for Research Use Only in accordance with applicable rules and regulations of the United States Food and Drug Administration and other applicable laws, and your sample shall not be used for patient care or diagnostic, clinical or therapeutic use. You agree to use CosmosID for Research Use Only. No analyses, reports, or other information obtained or provided through CosmosID are intended to be (nor should be relied upon as) medical advice or instructions for medical diagnosis or treatment.

CosmosID does not accept any responsibility for the accuracy of the input entered by the user or the consequences of any inaccuracies in this input. The analyses are not intended to replace professional medical care and attention by a qualified medical practitioner and consequently CosmosID does not accept any responsibility for the selection of drugs and the patient's response to treatment.

CosmosID is designed for whole genome shotgun (WGS) analysis on reads of at least 75 bases in length and should not be used for 16S analysis.



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY  
WASHINGTON, D.C. 20460

OFFICE OF CHEMICAL SAFETY  
AND POLLUTION PREVENTION

March 6, 2023

Brian Hogan  
Agent for Bluegreen US Water Technologies, Inc.  
c/o KRK Consulting LLC  
5807 Churchill Way  
Medina, O 44256

Subject: Labeling Notification per Pesticide Registration Notice (PRN) 98-10 – Delete statement related to shallow water  
Product Name: Lake Guard Oxy  
EPA Registration Number: 93647-2  
Application Date: 01/30/2023  
OPP Case Number: 00428733

Dear Mr. Hogan:

The U.S. Environmental Protection Agency (EPA) is in receipt of your application for notification under Pesticide Registration Notice (PRN) 98-10 for the above referenced product. The Biopesticides and Pollution Prevention Division (BPPD) has conducted a review of this request for its applicability under PRN 98-10 and finds that the action requested falls within the scope of PRN 98-10.

The labeling submitted with this application has been stamped “Notification” and will be placed in our records. You must submit one (1) copy of the final printed labeling with the modifications.

At this time, the U.S. Environmental Protection Agency (EPA) is not requiring a change to the restricted-entry interval (REI) of 0 hours (for non-spraying methods) and 1 hour (for spraying methods) as it appears on your labeling in the Agricultural Use Requirements Box. The EPA is currently developing guidance and criteria for when it will allow a REI of less than 4 hours and might require a change to the REI for this product in the future.

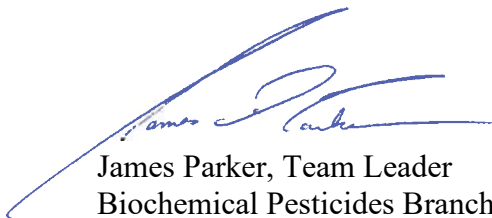
Should you wish to add/retain a reference to your company’s website on your label, then please be aware that the website becomes labeling under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and is subject to review by the EPA. If the website is false or misleading, the product will be considered to be misbranded and sale or distribution of the product is unlawful under FIFRA section 12(a)(1)(E). 40 CFR § 156.10(a)(5) lists examples of statements the EPA may consider false or misleading. In addition, regardless of whether a website is referenced on your product’s label, claims made on the website may not substantially differ from those claims approved through the registration process. Therefore, should the EPA find or if it is brought to our attention that a website contains false

Page 2 of 2  
EPA Reg. No. 93647-2  
OPP Case No. 00428733

or misleading statements or claims substantially differing from the EPA-approved registration, the website will be referred to the EPA's Office of Enforcement and Compliance Assurance.

If you have any questions, please contact Susannah Powell via email at [powell.susannah@epa.gov](mailto:powell.susannah@epa.gov).

Sincerely,

A handwritten signature in blue ink, appearing to read "James Parker", is written over a horizontal line.

James Parker, Team Leader  
Biochemical Pesticides Branch  
Biopesticides and Pollution  
Prevention Division (7511P)  
Office of Pesticide Programs



**NOTIFICATION**

93647-2

The applicant has certified that no changes, other than those reported to the Agency have been made to the labeling. The Agency acknowledges this notification by letter dated:

03/06/2023

# LAKE GUARD® Oxy

## Algaecide/Cyanobactericide

[Large Granules] [Small Granules] [Dust]

**ACTIVE INGREDIENT**

Sodium percarbonate..... 83.3%

**OTHER INGREDIENTS** ..... 16.7%

**TOTAL** ..... 100.0%

[UN 3378}

[CLASS 5.1]



Certified to  
NSF/ANSI/CAN 60

MUL 33 mg/L

### KEEP OUT OF REACH OF CHILDREN DANGER / PELIGRO

**Si usted no entiende la etiqueta, busque a alguien para que se la explique a usted en detalle.  
(If you do not understand the label, find someone to explain it to you in detail.)**

FIRST AID	
If in eyes	<ul style="list-style-type: none"> <li>• Hold eye open and rinse slowly and gently with water for 15-20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing.</li> <li>• Call a poison control center or doctor for treatment advice.</li> </ul>
If swallowed	<ul style="list-style-type: none"> <li>• Call a poison control center or doctor immediately for treatment advice.</li> <li>• Have person sip a glass of water if able to swallow.</li> <li>• Do not induce vomiting unless told to by a poison control center or doctor.</li> <li>• Do not give anything to an unconscious person.</li> </ul>
If on skin	<ul style="list-style-type: none"> <li>• Take off contaminated clothing.</li> <li>• Rinse skin immediately with plenty of water for 15-20 minutes.</li> <li>• Call a poison control center or doctor for treatment advice.</li> </ul>
If inhaled	<ul style="list-style-type: none"> <li>• Move person to fresh air.</li> <li>• If person is not breathing, call 911 or an ambulance, then give artificial respiration, preferably mouth-to-mouth if possible.</li> <li>• Call a poison control center or doctor for further treatment advice.</li> </ul>
Have the product container or label with you when calling a poison control center, doctor, or going for treatment. For non-emergency information concerning this product, call the National Pesticides Information Center (NPIC) at 1-800-858-7378 (NPIC Web site: <a href="http://www.npic.orst.edu">www.npic.orst.edu</a> ). For emergencies, call the poison control center 1-800-222-1222.	
NOTE TO PHYSICIAN: Probable mucosal damage may contraindicate the use of gastric lavage.	

EPA Reg. No. 93647-2

EPA Est. No. XXXXX-XX-X

Net Contents: 2, 5, 10, 20, 25, 50, 500, 1,000, 2,000, 2,204 lbs.

Batch Code:

**PRECAUTIONARY STATEMENTS**  
**Hazards to Humans and Domestic Animals**  
DANGER / PELIGRO

Corrosive. Causes irreversible eye damage and causes skin burns. May be fatal if swallowed. Harmful if absorbed through skin or inhaled. Do not get in eyes, on skin, or on clothing. Avoid breathing dust. Wear protective eyewear, such as goggles, face shield, or safety glasses. Wash thoroughly with soap and water after handling and before eating, drinking, chewing gum, using tobacco or using the toilet. Remove and wash contaminated clothing before reuse.

**Environmental Hazards**

This pesticide is toxic to birds. Do not apply this product or allow it to drift to blooming crops or weeds while pollinating insects are actively visiting the area.

**For container sizes 50 lbs. or greater:** Do not discharge effluent containing this product into lakes, streams, ponds, estuaries, oceans or other waters unless in accordance with the requirements of a National Pollutant Discharge Elimination System (NPDES) permit and the permitting authority has been notified in writing prior to discharge. Do not discharge effluent containing this product into sewer systems without previously notifying the local sewage treatment plant authority. For guidance contact your State Water Board or Regional Office of U.S. EPA. For additional information, refer to the product Material Safety Data Sheet.

**Physical and Chemical Hazards**

Oxidizing agent. Never use with other pesticides, cleaners, or oxidizing agents.

**Personal Protective Equipment**

Corrosive. Mixers, loaders, applicators, and other handlers must wear the following:

- Long-sleeved shirt and long pants
- Chemical resistant gloves
- Protective eyewear (goggles or face shield)
- Shoes plus socks

Follow manufacturer's instructions for cleaning/maintaining PPE. If no such instructions for washables exist, use detergent and hot water. Keep and wash PPE separately from other laundry.

**USER SAFETY RECOMMENDATIONS**

Users should remove clothing/PPE immediately if pesticide gets inside. Then wash thoroughly and put on clean clothing.

Users should remove PPE immediately after handling this product. Wash the outside of gloves before removing. As soon as possible, wash thoroughly and change into clean clothing.

**DIRECTIONS FOR USE**

It is a violation of Federal Law to use this product in a manner inconsistent with its labeling.

**GENERAL APPLICATION RESTRICTIONS**

Do not apply this product in a way that will contact workers or other persons, either directly or through drift. Only protected handlers may be in the area during application. For any requirements specific to your State or Tribe, consult the State or Tribal agency responsible for pesticide regulation.

This product is not intended as treatment against any public health organism for any use on this label. Do not apply directly to treated, finished drinking water reservoirs or drinking water receptacles when the water is intended for human consumption.

**SPRAY DRIFT MANAGEMENT**

A variety of factors including weather conditions (e.g., wind direction, wind speed, temperature, and relative humidity) and the method of application (e.g., ground or aerial) can influence pesticide drift. The applicator must evaluate all factors and make appropriate adjustments when applying this product.

### **Wind Speed**

Do not apply at wind speeds greater than 15 mph. Only apply this product if the wind direction favors on-target deposition (approximately 3 to 10 mph), and there are no sensitive areas within 250 feet downwind.

### **Temperature Inversions**

If applying at wind speeds less than 3 mph, the applicator must determine if a) conditions of temperature inversion exist, or b) stable atmospheric conditions exist at or below nozzle height. Do not make applications into areas of temperature inversions or stable atmospheric conditions.

### **Other State and Local Requirements**

Applicators must follow all state and local pesticide drift requirements regarding application of hydrogen peroxide compounds. Where states have more stringent regulations, they must be observed.

### **Equipment**

All aerial and ground application equipment must be properly maintained and calibrated using appropriate carriers or surrogates.

### **For aerial applications:**

The boom length must not exceed 75% of the wingspan or 90% of the rotor blade diameter. Release granules at the lowest height consistent with efficacy and flight safety. When applications are made with a crosswind, the swath must be displaced downwind. The applicator must compensate for this displacement at the up and downwind edge of the application area by adjusting the path of the aircraft upwind.

### **GENERAL INFORMATION**

Water bodies, such as impounded water, raw-water for drinking, lakes, ponds, reservoirs, waste water, irrigation, drainage, aquaculture, conveyance ditches, pipes, canals, laterals, estuaries, bayous, lagoons, and brackish and salt, sea and ocean water contaminated with algae/cyanobacteria can be treated with Lake Guard® Oxy.

Do not apply when wind speed favors drift beyond the area intended for treatment.

~~Do not apply to shallow water bodies less than three feet deep, or to areas where surface water is stagnant.~~ Do not use the product to treat ornamental fish (i.e., fish that grow in home aquariums or outdoor tanks for aesthetic purposes).

Water hardness, temperature of the water, type and cell density of algae/cyanobacteria to be controlled, and the amount of water flow are to be considered in using Lake Guard® Oxy to control algae/cyanobacteria. Treated water resource should be monitored systematically for the presence of harmful algae/cyanobacteria using adequate apparatuses. Begin treatment as soon as algal/cyanobacterial cell numbers reach 5,000-20,000 cells/mL (and below 10 µg chlorophyll-a/liter).

If treatment is delayed until algal/cyanobacterial cell numbers exceed 20,000 cell/mL (or equivalently, above 10 µg chlorophyll-a/liter), an increase in the quantities of Lake Guard® Oxy will be required, as well as in treatment frequency. Use caution when treating heavy blooms, as oxygen loss from mass decomposition of dead algae/cyanobacteria can cause fish suffocation. See application rates section for treating heaving blooms.

Always apply the granules off wind and let the wind and currents to carry them where algae/cyanobacteria cell masses are concentrated. It is best to treat algae on a sunny day at morning hours. If there is some doubt about the amount of Lake Guard® Oxy to apply, it is best to start with the lower application rate. The quick and easy treatment application (minutes for applying a hundred lb), and

the visual results that would be apparent within 48-72 hours, enable the applicator to make a quick adjustment on whether to increase the dose rate by 50-100% during the next treatment application, or scale it back and use even less product under similar conditions. Since all water bodies are unique aquatic ecosystems, and, therefore, differ from each other in treatment response and longevity, the Lake Guard® Oxy application protocol offers the applicator a scalable dose rate that saves money and reduces the chemical load introduced to the environment. For first time users, or when in doubt, applicators should start with 0.5-5 lb/acre and adjust the dose rates (increase or decrease) by increments of 25-50%, and over time can fine-tune the treatment application for each waterbody.

## **APPLICATION RATES**

The best method by which to apply Lake Guard® Oxy granules to water is by broadcasting (dusting) it over a well-defined contamination zone, at early bloom stages, when harmful algal/cyanobacteria numbers are at 5,000 to 20,000 cells/mL (and below 10 µg chlorophyll-a/liter).

### Determination of surface area to be treated

Determine the size of the infested area as follows: (1) in small infested reservoirs, under than 250 acres, obtain surface area by measuring of regular shaped ponds or mapping of irregular ponds or by reference to previously recorded engineering data or maps. (2) In water bodies larger than 250 acres (or smaller ponds with a defined contaminated zone) outline the infested area by a combination of the following instruments: microscopical count, pigment extraction, toxin evaluation, probes that detect specific pigments that are known to serve as a correlated proxy for algae/cyanobacteria biomass, satellite imaging, etc. NOTE: evaluation of the state of the infestation should be done by professional personnel.

### Determination of the application rate

For control of harmful algae/cyanobacteria infestation it is essential to begin Lake Guard® Oxy treatment when harmful algae/cyanobacteria cell numbers are in the range of 5,000-20,000 cell/mL (or below 10 µg chlorophyll-a/liter). Apply 0.5-5 lbs./acre Lake Guard® Oxy at these algal/cyanobacterial cell-densities. Always start with the lower rate. At higher infestation rates, when cyanobacterial cell density is between 20,000-100,000 cells/mL (or between 10-50 µg chlorophyll-a/liter) use 5-30lbs./acre Lake Guard® Oxy.

If treatment is delayed until algal/cyanobacterial cell numbers exceed 100,000 cell/mL (or equivalently, above 50 µg chlorophyll-a/liter), an increase in the quantities of the Lake Guard® Oxy will be required, as well as in treatment frequency. Therefore, in heavy blooms, when cyanobacterial scum or aggregates are visible to the naked eye (more than 100,000 cells/mL of algae/cyanobacteria or over 50 µg chlorophyll-a/liter), treat with doses between 30-98 lbs./acre. If doses exceed 98 lbs./acre, treat no more than one-half of the water area in a single application. Maximum single application rate allowed should not exceed 294 lbs./acre of the Lake Guard® Oxy. NOTE: when cyanobacterial aggregated could be seen with the naked eye, the cyanobacterial cell density in the water is estimated to exceed 100,000 cells per ml.

When a single application dose is below 30 lbs./acre, minimum retreatment interval is 12 hours. When a single application dose is between 30-98 lbs./acre, minimum retreatment interval is 24 hours. When a single application dose exceeds 98 lbs./acre, the minimum retreatment interval is 48 hours.

## **APPLICATION METHOD**

Apply Lake Guard® Oxy using equipment designed for granular dusting. Dusting can be done manually by hand or by a boat or airplane, depending on the area of the zone and its proximity to reservoir's bank. When a small duster is mounted on a properly equipped boat, application can be broadcast directly on the water surface at the edge of the infested zone. Note that the direction of the wind is an important factor - always dust off-wind. Do not use this method unless completely familiar with this type of application.

## STORAGE AND DISPOSAL

Do not contaminate water, food, or feed by storage or disposal.

**PESTICIDE STORAGE:** Keep pesticide in original container. Do not use in food or drink containers.

**PESTICIDE DISPOSAL:** Pesticide wastes may be hazardous. Improper disposal of excess pesticide, spray, mixture or rinsate is a violation of Federal Law. If these wastes cannot be disposed of by use according to label instructions, contact your State Pesticide or Environmental Control Agency, or the Hazardous Waste Representative at the nearest EPA Regional Office for guidance.

**CONTAINER HANDLING:** Nonrefillable container. Do not reuse or refill this container. Completely empty bag into application equipment, then offer for recycling if available or dispose of empty bag in a sanitary landfill or by incineration.



## CONDITIONS OF SALE AND LIMITATIONS OF WARRANTY AND LIABILITY

Read the entire directions for use, conditions of warranties and limitations of liability before using this product. If terms are not acceptable, return the unopened product container at once. By using this product, user or buyer accepts the following Conditions, Disclaimer of Warranties and Limitations of Liability. **CONDITIONS:** The directions for use of this product are believed to be adequate and must be followed carefully. However, it is impossible to eliminate all risks associated with the use of this product. Ineffectiveness or other unintended consequences may result because of such factors as weather conditions, presence of other materials, or the manner of use or application, all of which are beyond the control of BlueGreen Water Technologies. All such risks shall be assumed by the user or buyer.

**DISCLAIMER OF WARRANTIES:** To the extent consistent with applicable law, BlueGreen Water Technologies makes no other warranties, express or implied, of merchantability or of fitness for a particular purpose or otherwise, that extend beyond the statements made on this label. No agent of BlueGreen Water Technologies is authorized to make any warranties beyond those contained herein or to modify the warranties contained herein. To the extent consistent with applicable law, BlueGreen Water Technologies disclaims any liability whatsoever for special, incidental or consequential damages resulting from the use or handling of this product. **LIMITATIONS OF LIABILITY:** To the extent consistent with applicable law, the exclusive remedy of the user or buyer for any and all losses, injuries or damages resulting from the use or handling of this product, whether in contract, warranty, tort, negligence, strict liability or otherwise, shall not exceed the purchase price paid or at BlueGreen Water Technologies' election, the replacement of product.

Property rights are protected under the patent legislation of - USA Patent No. 10,729,138 and USA Patent No. 10, 092, 005; Indian Patent no. 201617001647; Mexican Patent No. MX/a/2016/000199; South African Patent No. 2016/00478; the Russian Federation Patent No. 2687929.

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**GOAL** - BlueGreen Water Technologies (BlueGreen) is seeking partners for strategic projects to restore large-scale water bodies. Our core focus is long-term remediation of impaired water quality.

**COMPANY** - BlueGreen is a research-development-commercialization company focused on large scale solutions to improving water quality, as well as remediating harmful algal blooms and their underlying causes. The U.S. company is headquartered in Florida, with corporate headquarters located in Israel. To date, we have multiple documented successes around the world, including the state of Florida, using novel approaches. These documents can be provided upon request.

**TECHNOLOGY** - BlueGreen developed two core technologies to introduce for strategic projects:

- *Treatment Technology:* our treatment products induce natural Programmed/Regulated Cell Death (PCD/RCD) via a floating, time-release methodology to the top layer of the water body. The floating, time-release treatment follows the colonial aggregates of harmful algae via the same wind and water currents carrying the bloom and enable advanced efficacy with minimal doses of products. However, the benefits of our products do not stop at only treating harmful algae blooms. Two of our three products will also target nutrient management through catalytic chemical reactions (when using our hydrogen peroxide-based formula) and coagulation (when using our Alum-based formula).
- *Monitoring Analytics:* our algorithms offer advanced satellite, drone and in-situ monitoring that can identify leading indicators and track blooms; as our AI “learns” the unique characteristics of the water, we enable modeling and forecasting in ways far more advanced than anything being done in this area of study.

**SCALE AND FOCUS** - Our tech is oriented toward “any scale” water bodies, such as impounded water, raw-water for drinking, lakes, ponds, reservoirs, wastewater, irrigation, drainage, aquaculture, conveyance ditches, pipes, canals, laterals, estuaries, bayous, lagoons, and brackish and salt, sea and ocean water contaminated with algae/cyanobacteria can be treated with Lake Guard® Oxy.

**PAST PERFORMANCE** - BlueGreen has a proven track record of rehabilitating HAB infested water bodies around the world, including USA, Africa, China, and Israel, and will soon enter markets in Colombia and Ecuador.

**CURRENT PROJECT OF NOTE** - We’re working with a renowned HAB/Water Quality expert (Dr. Hans Paerl) from University of North Carolina-Chapel Hill on an efficacy study at Lake Mattamuskeet (40,000 acres), a well-known U.S. Fish and Wildlife National Wildlife Refuge and pocosin lake in North Carolina.

**ADDED BENEFIT: CARBON CAPTURE** - BlueGreen has proven and are currently socializing our results for massive levels of carbon capture and sequestration as a byproduct of our treatment methodology. In short, by timing the treatments in alignment with the bloom’s growth phases, we

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are able to capture huge amounts of carbon from the atmosphere and sequester as biomass to induce biomineralization (lime).

**GOAL ALIGNMENT OUTLINE:**

BlueGreen is committed to playing a pivotal role in restoring balance in natural water bodies around the world. Toxic harmful algal blooms (HABs) are increasing in both magnitude and frequency due to a variety of human factors, catalyzed by excessive nutrient loading upon which the harmful algae feed. By the time blooms forms, the system is already severely out of balance and requires intervention via a combination of long-term and near-term efforts.

BlueGreen’s solutions include both advanced monitoring and environmentally friendly treatments suitable for the most sensitive ecosystems. While addressing excessive nutrient loading takes decades, BlueGreen offers an optimal path to safely accelerate results and monitor progress based on the following core values:

- Habitat Restoration occurs through our nature-based, targeted treatments against invasive, chronic, and/or toxic colonies of harmful algae. Our treatment process specifically triggers “Programmed Cell Death” (PCD; also known as Regulated Cell Death or Apoptosis) in cyanobacteria through temporary upper-layer oxidative stress while preserving an underwater wildlife habitable zone during the treatment phase. Current scientific research is exploring the phenomenon across other phytoplankton species as well. Nevertheless, upon collapse of the invasive cyanobacteria colony, other beneficial phytoplankton quickly begin to fill the ecological niche and begin competing for nutrient sources. This enhanced biodiversity quickly accelerates the healing process for the water to gain a more proper balance in the ecosystem, which is a critical element of on which Habitat Restoration depends.
- Leave No Trace project methodologies, that require no permanent infrastructure, utilities, or equipment, preserves (and enhances) an ecosystem in its natural state after our treatments. We perform pre-treatment and ongoing water analysis through advanced satellite-based monitoring; when treatments are needed, our field presence is temporary in nature in that our activities create the least amount of disturbance to the system as possible – lasting only 1 or 2 days per treatment.
- Water Quality is enhanced through our remediation efforts against harmful algal blooms and their underlying causes. By resetting the balance of the ecosystem through intervention that selectively targets cyanobacteria colonies, we are specifically and unequivocally reducing the propensity for water toxicity that results from HABs, thus accelerating and enabling further restoration.
- Climate Solutions are an added benefit to the methodologies developed by BlueGreen. By studying the behaviors of the cyanobacteria colonies in water and devising treatment plans that trigger PCD, collapsing colonies absorb immense amounts of carbon from the atmosphere as their biomass sinks to begins a biomineralization process within the sediment.

**SOLUTION PORTFOLIO:**

BlueGreen leads the market as the only technology-driven developer of holistic HAB Management solutions designed for unlimited scale. Our products, services and resources are fully integrated to enable monitoring, data capture, treatment, and sustainment against harmful algae blooms.

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- *Lake Guard View*<sup>®</sup> - Advanced AI-Based Monitoring and Analysis Platform
  - Autonomous Remote Monitoring via daily satellite imagery analytics
  - Autonomous In-Situ remote monitoring with near real-time analytics
  - Advanced ad-hoc analysis via Drone-enabled app analytics
  - Treatment planning and modelling dashboards
- *Lake Guard Oxy*<sup>®</sup> preventative and bloom-stage water treatment
  - Fully biodegradable, peroxide-based formulation
  - Floating time-release mechanism to induce oxidative-stress PCD
  - Shoreline, boat or aerial application
- *Lake Guard Blue*<sup>®</sup> preventative and bloom-stage water treatment
  - Copper sulphate-based active ingredient
  - Floating time-release delivery mechanism for optimized efficacy against HABs
  - Shoreline, boat or aerial application
- Project & Logistics Management, Field / On-Water Services, and Emergency Response
  - Logistics design for project management and emergency response
  - Licensed applicators
  - Water-sample collection and analysis
  - On-call services
- Water Science Services
  - Science as a Service, Training and Consultation
  - Long-term rehabilitation design and management
  - Research and development for unique scenarios

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**JESSICA R. FROST**

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Social Media: [https://www.researchgate.net/profile/Jessica\\_Frost](https://www.researchgate.net/profile/Jessica_Frost)

***EDUCATION***

Ph.D. Biological Oceanography, 2014

Institut für Hydrobiologie und Fischereiwissenschaft, Universität Hamburg, Germany

Dissertation: *Gelatinous Zooplankton Trophodynamics: Ecological and Biogeochemical Influences on Pelagic Food Webs*

Advisors: Drs. Clive J. Fox, Marsh J. Youngbluth, Michael A. St. John, and Myron A. Peck

Disputation Judges: Drs. Friedrich Buchholz, Aino Hosia, Cornelia Jaspers, Rolf Koppelman, Myron Peck, Delphine Thibault, and Justus van Beusekom

Disputation Moderator: Dr. Christian Möllmann

M.Sc. Fisheries and Aquatic Sciences, 2003

Department of Fisheries and Aquatic Sciences, University of Florida, USA

Thesis: *Ecology of Lake Griffin, a hypereutrophic cyanobacteria dominated lake in central Florida*

Advisor: Dr. Edward J. Philips Committee: Drs. Charles Cichra and Mark Brenner

B.Sc. Environmental Science, Technology, 2001

College of Natural Resources and Environment, University of Florida, USA

Dean: Dr. Stephen R. Humphrey

***RESEARCH INTERESTS***

I am broadly interested in freshwater and marine ecology, with emphasis on factors influencing indicator species, community assemblage, biogeochemical cycling, water quality, and trophic state. Much of my career has centered on water quality, ecological interactions and potential correlations and linkages.

***PROFESSIONAL EXPERIENCE***

***(A) Full-Time 40 hr Positions***

<b><u>Date/Salary</u></b>	<b><u>Employer</u></b>	<b><u>Position &amp; Selected Duties</u></b>
11/2022-present	<b>BlueGreen Water Technologies</b>	Scientific Director (USA)
12/2019-02/2022	<b>South Florida Water Management District</b>	Section Administrator, Coastal Ecosystems Supervise 18 staff scientists (biologists & modelers); Conduct & manage monitoring research regarding ecology of estuaries, Florida

		<p>Statute minimum flows &amp; minimum levels, CERP RECOVER indicator species, &amp; Biscayne Bay agricultural engineering; Procurement, Budget management  Emergency Operations Planning Section Chief;  Loxahatchee River Management Coordination Council Member  BGA Task Force Member  LOSOM PDT Member  BBSEER PDT Member</p>
07/2010-12/2019	<b>University of Florida</b>	<p>Laboratory Manager/Research Coordinator/  Post Doc  Conduct &amp; manage sponsored research; Staff supervisor; FLDACS permitting; Graduate student mentor; Field sampling; Wet chemistry analyses; Budget, data, boat, fleet, equipment, &amp; lab management; Data analysis; Manuscript &amp; grant writing</p>
01/2006-04/2009	<b>Universität Hamburg</b>	<p>PhD fellowship; Eur-Oceans Network of Excellence</p>
04-12/2005	<b>Microbac Laboratories, Inc.</b>	<p>Chemist  Laboratory analyses, quality control &amp; quality assurance, SOP writing, flow injection auto analyzer installation</p>
08-12/2004	<b>Harbor Branch  Oceanographic Institution</b>	<p>Research Associate  Cruise logistics, laboratory analyses, data management, &amp; manuscript writing</p>
05/2003-08/2004	<b>University of Florida</b>	<p>Research Assistant  Field sampling, laboratory analyses, data management &amp; analysis, manuscript writing</p>
01/2001-05/2003	<b>University of Florida</b>	<p>Graduate Research Assistant  Field sampling, laboratory analyses, data management &amp; analysis, &amp; manuscript writing</p>
01/1998-05/2001	<b>University of Florida</b>	<p>Laboratory Technician  Laboratory analyses, data management</p>

**(B) Part-time as needed Positions**

2011	Sea Pen Scientific Writing LLC Owner: Dr. Jennifer Purcell	Editing and Research Services edit manuscripts for submission to peer-reviewed journals
05-07/2009	DTU Aqua, Denmark	Visiting scientist (host: Dr. Thomas Kiørboe), manuscript preparation

**(B) Teaching**

<b>Date</b>	<b>Employer</b>	<b>Course</b>
2015	University of Florida	Seminar in Fisheries & Aquatic Sciences FAS 4933
2006	Universität Hamburg	Trophodynamics Interactions 14.684
2001	University of Florida	Fish and Limnology FAS 6932

**(C) Grant Writing**

<b>Date</b>	<b>Call</b>	<b>Title</b>	<b>Budget</b>
2015	Florida Sea Grant Statement of Interest 2016-2017	Development of New Broodstock for Reintroduction of Hard Clam ( <i>Mercenaria mercenaria</i> ) Aquaculture and Restoration of Environmental Condition in the Indian River Lagoon	\$198, 000
2014	University of Florida IFAS Research Equipment and Infrastructure Funding 2014	FlowCAM	\$75, 000
2014	University of Florida 2014 Early Career Scientist Seed Funding	Jellyfish as Bioindicators	\$82, 840

**(D) Publications**

22. *Water and Climate Resilience Metrics*, PHASE:1 LONG-TERM OBSERVED TRENDS. Final Report December 17, 2021. South Florida Water Management District.
21. Detong Sun, **Jessica Frost**, Mark Barton, Melanie Parker and Y. Peter Sheng (submission). Estuarine Water Quality: One-Dimensional Model Theory and Its Application to a Riverine Subtropical Estuary in Florida. Target journal *Estuaries & Coasts*.
20. **Frost, J.R.** (in prep). Fine-scale behavior of three developmental stages of *Acartia tonsa* Dana, 1849 (Copepoda: Calanoida) relative to salinity gradients and thin layers of phytoplankton.

19. **J.R. Frost**, C.A. Jacoby, T.K. Frazer (in prep). Mat manipulation of *Lyngbya sp* from a spring-fed system in Florida: Growth potential and biogeochemical effects.
18. Jacoby, C.A., **J.R. Frost**, M.J. Youngbluth, T.K. Frazer (in prep). Distribution, abundance, oxygen consumption and predatory impact of *Beroe cucumis* and *Bolinopsis infundibulum* (Ctenophora) in the Gulf of Maine and Oceanographer Canyon.
17. Lebrato, M., M. Pahlow, **J. Frost**, M. Küter, P. de Jesus Mendes, J-C. Molinero, A. Oschiles (2019). Sinking of Gelatinous Zooplankton Biomass Increases Deep Carbon Transfer Efficiency Globally. *Global Biogeochemical Cycles*. Vol. 33. Issue 12. Pg. 1764-1783.
16. Hyman, A., T. Frazer, C. Jacoby, **J. Frost**, M. Kowalewski (2019). Long-term persistence of structured habitats: Seagrass meadows as enduring hotspots of biodiversity and faunal stability. *Proceedings of the Royal Society B*. 286: 20191861.
15. **J.R. Frost** (2017). Living in a Spineless World: Jellyfish Life. Florida LAKEWATCH Newsletters vol. 78, pp 6–8. Extension publication.
14. Jacoby, C.A., T.K. Frazer, M.A. Edwards, **J.R. Frost** (2015). Water quality characteristics of the nearshore Gulf coast waters adjacent to Hernando, Citrus and Levy Counties. Project COAST 1997-2014. Final Report. Southwest Florida Water Management District, Brooksville, Florida. 24 pp. (Contract#09C00000052; Project B678)
13. Jacoby, C.A., T.K. Frazer, **J.R. Frost**, L.G. Chambers, S.K. Notestein (2013). Kings Bay sediment feasibility study. Annual Report. Southwest Florida Water Management District, Brooksville, Florida. 8 pp. (Contract#W472)
12. **Frost, J.R.**, C.A. Jacoby, T.K. Frazer, A.R. Zimmerman (2012). Pulse perturbations from bacterial decomposition of *Chrysaora quinquecirrha* (Scyphozoa: Pelagiidae). *Hydrobiologia* 690: 247–256.
11. **Frost, J.R.**, A. Denda, C.J. Fox, C.A. Jacoby, R. Koppelman, M. Holtegaard Nielsen, M.J. Youngbluth (2012). Distribution and trophic links of gelatinous zooplankton on Dogger Bank, North Sea. *Marine Biology* 159: 239–253.
10. Robinson, C. *et al.* (2010). Mesopelagic zone ecology and biogeochemistry – a synthesis. *Deep-Sea Research II* 57 (16): 1504–1518.
9. **Frost, J.R.**, C.A. Jacoby, M.J. Youngbluth (2010). Behavior of *Nemopsis bachei* L. Agassiz, 1849 in the presence of physical gradients and biological thin layers. *Hydrobiologia* 645: 97–111.
8. Jacoby, C.A., M.J. Youngbluth, **J.R. Frost**, P.R. Flood, F. Uiblein, U. Bämstedt, F. Pagès, D. Shale (2009). Vertical distribution, behavior, chemical composition and

metabolism of *Stauroteuthis syrtensis* (Octopoda: Cirrata) in the northwest Atlantic. *Aquatic Biology* 5: 13–22.

7. **Frost, J.R.**, E.J. Phlips, R.S. Fulton III, C.L. Schelske, W. Kenney, M. Cichra (2008). Temporal trends of trophic state variables in a shallow hypereutrophic subtropical lake, Lake Griffin, Florida, USA. *Fundamental and Applied Limnology (Archiv für Hydrobiologie)* 172 (4): 263–271.
6. Koppelman, R., **J. Frost** (2008). The ecological role of zooplankton in the twilight and dark zones of the ocean, In: *Biological Oceanography Research Trends*, ed. Mertens, LP, Nova Science Publishers, Inc., New York, pp. 67–130.
5. Christiansen, B. *et al.* (2007). The ecology and biogeochemistry of Anaximenes Mountains, Cruise No. M71, Leg 1, 11-24 December 2006, Heraklion, Meteor-Berichte. 50 pp.
4. **Frost, J.** (2006). Gelatinous zooplankton: evolution of sampling techniques. *Eur-Oceans Newsletter* 3. [www.eur-oceans.eu](http://www.eur-oceans.eu)
3. Phlips, E.J., **J. Frost**, M. Yilmaz, N. Steigerwalt, M. Cichra (2004). Factors controlling the abundance and composition of blue-green algae in Lake Griffin. Final Report to the St. Johns River Water Management District, Palatka, Florida. (Project #SF669AA)
2. Phlips, E.J., M. Cichra, **J. Frost** (2002). Assessment of Lake Griffin algal blooms. Final Report to the St. Johns River Water Management District, Palatka, Florida. (Contract#SD419AA)
1. Phlips, E.J., E. Bledsoe, M. Cichra, S. Badylak, **J. Frost** (2002). The distribution of potentially toxic cyanobacteria in Florida, p. 22–29. In Monograph: “Proceedings of Health Effects of Exposure to Cyanobacteria Toxins: State of the Science,” August 13–14, 2002.  
<http://www.doh.state.fl.us/Environment/community/aquatic/cyanobacteria.htm>

### **(E) Presentations**

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13. **Jessica Frost** (2021). Seagrass. SFWMD Governing Board Member Invited Oral Presentation Town of Sewall’s Point Commission Meeting, December 14, 2021.
12. **Frost, J.** (2018). Professional TED Talk: Water Quality. Invited Oral Presentation Institute of Food and Agricultural Sciences, School of Forest Resources and Conservation, Fisheries and Aquatic Sciences Graduate Student Symposium, University of Florida, Gainesville, Florida, February 16, 2018.
11. Frazer, T., M. Kowalewski, **J. Frost** (2017). Historical Ecology of Seagrass Meadows: Assessing Multi-Centennial Dynamics of Threatened Biodiversity Hotspots. Poster

Presentation Institute of Food and Agricultural Sciences Symposium, University of Florida, Gainesville, Florida, May 2017.

10. Jacoby, C.A., **J.R. Frost**, M.J. Youngbluth, T.K. Frazer (2013). Distribution, abundance, oxygen consumption and predatory impact of *Beroe cucumis* and *Bolinopsis infundibulum* (Ctenophora) in the Gulf of Maine and Oceanographer Canyon. Oral Presentation Fourth International Jellyfish Blooms Symposium, Hiroshima, Japan, June 5–7, 2013.
9. **Frost, J.R.**, C.A. Jacoby, T.K. Frazer, A.R. Zimmerman (2012). Pulse perturbations from bacterial decomposition of *Chrysaora quinquecirrha* Desor, 1848 (Scypozoa: Pelagiidae). Oral Presentation UF Marine Biology Symposium, Whitney Marine Laboratory, St. Augustine, Florida, January 19–20, 2012.
8. **Frost, J.R.**, C.A. Jacoby (2011). Jellyfish and chips. The next “It’s What’s For Dinner” in Florida? Invited Poster Presentation CERF 2011: Societies, Estuaries & Coasts: Adapting to Change, Daytona Beach, Florida, November 6–10, 2011.
7. **Frost, J.R.**, C.A. Jacoby, M.J. Youngbluth (2010). Behavior of *Nemopsis bachei* L. Agassiz, 1849 in the presence of physical gradients and biological thin layers. Poster Presentation ASLO Ocean Sciences Meeting, Portland, Oregon, February 22-26, 2010.
6. **Frost, J.R.** (2009). Interactions between gelatinous zooplankton and discontinuities: Influence on pelagic food webs. Oral Presentation Technical University of Denmark (DTU Aqua Kaffeklubben), Charlottenlund, Denmark. June 24, 2009.
5. **Frost, J.R.**, M.A. St. John, M.J. Youngbluth, C.J. Fox (2008). Distribution and trophic role of gelatinous zooplankton over the frontal system of Dogger Bank, North Sea. Oral Presentation Eur-Oceans Final Meeting, Rome, Italy. November 25-27, 2008.
4. **Frost, J.R.**, C.A. Jacoby, G. Gust, M.H. Nielsen, R.W. Campbell, M.A. St. John (2007). Behavioral and biochemical effects of hydrostatic pressure changes on *Acartia tonsa* (Dana, 1848) (Copepoda: Calanoida): A methodological approach. Oral Presentation PICES 4<sup>th</sup> International Zooplankton Production Symposium, Hiroshima, Japan. May 28-June 1, 2007.
3. **Frost, J.R.**, M. St. John, C. Fox, M. Youngbluth (2006). Biological, chemical and physical interactions within thin layers: Resolving the trophic role of gelatinous zooplankton. Poster Presentation Eur-Oceans/NATO ‘Climate Change Impacts on Marine Ecosystems’ Summer School, Ankara, Turkey. August 14-26, 2006.
2. Youngbluth, M.J., C.A. Jacoby, **J.R. Frost**, D. Shale (2005). Distribution, metabolism and behavior of the deep-living cephalopod *Stauroteuthis syrtensis* (Suborder Cirrata). Oral Presentation ASLO International Meeting, Santiago de Compostela, Spain. June 19-24, 2005.

1. **Frost, J.R.,** E.J. Phlips, M.F. Cichra (2003). Ecology of Lake Griffin, a hypereutrophic cyanobacteria-dominated lake in central Florida. Oral Presentation NALMS 23<sup>rd</sup> Annual International Symposium, Mashantucket, Connecticut, USA. November 5-7, 2003.

## AWARDS

<b>Date</b>	<b>Award</b>
2022	South Florida Water Management District 2021 Team of the Year: <i>Water Reservations Team- EAA Reservoir and Kissimmee</i>
2021	South Florida Water Management District April 2021 Team of the Month: <i>Kissimmee Water Reservation Team</i>
2021	South Florida Water Management District February 2021 Team of the Month: <i>EAA Reservoir Water Reservations Team</i>
2013	COSEE Florida BioCube educator workshop – Biocube: exploring backyard diversity
2011	MESOAQUA workshop – Mesocosms in aquatic ecology: use, problems and potentials
2006-09	Eur-Oceans PhD Fellowship Award (WP4-SYSMS-1101; Director: Dr. Carlos Duarte); European Union Network of Excellence
2006	Scholarship to the Eur-Oceans/NATO “Climate Change Impacts on Marine Ecosystems” Summer School
2003	North American Lake Management Society (NALMS) Best Student Paper Winner
2002	Florida Certificate of Boating Education

## SKILLS

<b>Laboratory:</b>	certified QA/QC chemical analyst	nutrient enrichment bioassays	
	algal culture	spectrophotometry	
	<i>in situ</i> data logging	gas chromatography	
	Winkler titrations	fluorometry	
	inverted phase-contrast microscopy	biochemical oxygen demand	
	stable isotopes	wet chemistry	
	Beckman Coulter Counter	bacterial enumeration	
	fiber optic oxygen meter (respirometry)		
	<b>Taxonomy:</b>	cyanobacteria	phytoplankton
		Cnidaria & Ctenophora	mesozooplankton
Vascular/non-vascular plants (seagrasses/macroalgae)			

**Field sampling:** phytoplankton submerged aquatic plants  
zooplankton benthic invertebrates  
fish water quality monitoring  
sediments

**Field gear:** MOCNESS WP2 net  
CTD Van Dorn  
Integrated Pole Purse Sein  
Otter Trawl Tucker Trawl  
Long Line Manned Submersible  
Video Plankton Recorder (VPR) Remotely Operated Vehicle (ROV)  
SCUBA with NITROX Boats ( $\leq 28$  ft.)  
Li-Cor datalogger YSI datalogger  
Sieves/Floating Sieves PVC Quadrats  
Modified Venturi vacuum pump w/generator

**Software:** Microsoft Office SAS Procite MatLab ArcGIS 10.1 Minitab 17

**Languages:** English (Native)  
Spanish (CEFR C1 level of proficiency)  
German (CEFR B1 level of proficiency)

**Permitting:** Florida Department of Agriculture and Consumer Services – Nursery Registration

**OTHER PROFESSIONAL ACTIVITIES**

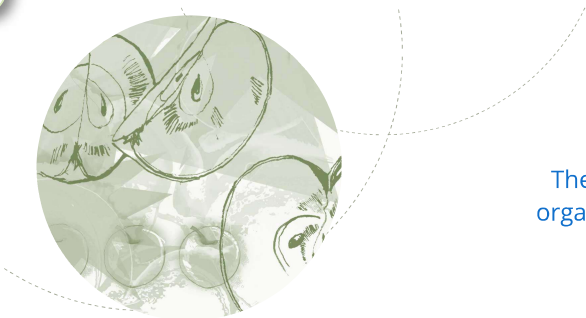
<b>Date</b>	<b>Activity</b>	<b>Duties</b>
2022	Loxahatchee River Management Coordination Council	SFWMD Representative
2021	SFWMD Blue Green Algae Task Force: Led by Rory Feeney	Participant
2021-22	Cross-functional QA Team (led by WQB)	Participant
2021-22	BBSEER partnership with USACE	SFWMD PDT
2021	Seagrass-with SFWMD Office of Communications and Kimberly Miller of Palm Beach Post	Press Interview
2020-22	FDEP Coastal Managers Forum	SFWMD Representative
2020-22	LOSOM partnership with USACE	SFWMD PDT
2020-22	ASB Point of Contact for Field Operations Center	Mediator/Communication



2020-22	SFWMD EOC	Planning Chief
2020-22	CHNEP Technical Advisory Committee	SFWMD Back-up
2020-22	IRL HAB Coordination	SFWMD Representative
2020-22	SFWMD COVID Strike Force Team	ASB Representative
2020-22	IRLNEP Grant Program	Reviewer
2020-21	Indian River Lagoon Symposium	Participant
2019-22	SFWMD Weekly Operations Meeting	ASB co-Host
2019-21	SFWMD SFER Vol. II Chapters 5B, 8C & 8D	Reviewer/Editor
2020	EPA Competitive Grant Program: Approaches to reduce nutrient loadings for harmful algal blooms management Announcement Number: EPA-G2020-STAR-A1	Reviewer (paid)
2017	<i>Nature Research</i> Scientific Reports	Reviewer
2017	Tampa Bay Watch: harvest plugs of seashore paspalum from Port Manatee, October 11, 2017	Volunteer
2017	Deutschlandradio (German Public Radio Station) Dagmar Röhrlich with Deutschlandfunk - piece on abilities and significance of jellyfish in oceans, food webs and ecosystems	Press Interview
2017	<i>Marine Ecological Processes</i>	Reviewer
2016	<i>Journal of Fisheries and Aquaculture Research</i>	Reviewer
2016	Ctenopalooza: an international meeting of ctenophore biologists- University of Florida Whitney Laboratory	Participant
2015	USGS/Save Crystal River: Population Status of Kings Bay Manatees (health assessment)	Volunteer
2015	stemCONNECT program of Florida High Tech Corridor Council	Participant
2015	Oregon Sea Grant	Reviewer
2015	Indian River Lagoon Symposium 2015: Lessons, Challenges, and Opportunities- Ft. Pierce, Florida	Participant
2014-present	<i>Journal of Plankton Research</i>	Reviewer
2014-present	<i>Hydrobiologia</i>	Reviewer
2014	National Science Foundation Division of Ocean Sciences Biological Oceanography Program	Reviewer

2014	ICES Annual Science Meeting- A Coruña, Spain	Participant
2014-present	<i>Journal of Experimental Marine Biology and Ecology</i>	Reviewer
2014-present	<i>Water Research</i>	Reviewer
2013	UF & Florida Sea Grant ArcGIS 10.1 workshop	Participant
2013	COSEE Florida BioCube educator workshop – BioCube – exploring backyard diversity- St. Teresa, Florida	Participant
2012	Indian River Lagoon Symposium 2012: Looking Forward- Ft. Pierce, Florida	Participant
2012-present	<i>Fisheries Research</i>	Reviewer
2011	2 <sup>nd</sup> DTU Aqua Jelly Day- <i>Mnemiopsis leidyi</i> in European Waters: Where are they and what do we know?- Charlottenlund, Denmark	Participant
2011	MESOAQUA workshop – Mesocosms in aquatic ecology: use, problems and potentials- Kiel, Germany	Participant
2011	Guest co-editor <i>Hydrobiologia</i>	Edited manuscripts for special volume
2011-present	<i>Invertebrate Biology</i>	Reviewer
2011-present	<i>Journal of the Marine Biological Association of the United Kingdom</i>	Reviewer
2010-present	<i>Chinese Journal of Oceanology and Limnology</i>	Reviewer
2010	Third International Jellyfish Blooms Symposium- Mar del Plata, Argentina	Participant
2010	ASLO Ocean Sciences Meeting- Portland, Oregon	Presenter
2008-present	<i>Marine Biology</i>	Reviewer
2008	IMBER IMBIZO Workshop “Integrating biogeochemistry and ecosystems in a changing ocean”- Miami, Florida	Participant, Co-author summary paper published in <i>Deep-Sea Research II</i>
2008	FS Maria S. Merian (MSM 07/3)	Ecological assessment of gelatinous

2007	FS Alkor (AL300)	zooplankton, Walvis Bay, Namibia Ecological assessment of gelatinous zooplankton, Dogger Bank, North Sea
2006	FS Meteor (M71/1)	Ecological assessment of gelatinous zooplankton, Anaximander Seamount, Mediterranean Sea
2006	Eur-Oceans/NATO Summer School & Advanced Study Institute "Climate Change Impacts on Marine Ecosystems"- Ankara, Turkey	Participant, Presenter
2005	ASLO International Meeting- Santiago de Compostela, Spain	Participant, Co-author
2005	R/V Seward Johnson I	Midwater sampling using submersibles
2004	R/V Seward Johnson II	Midwater sampling using submersibles
2003	North American Lake Management Society Symposium- Mashantucket, Connecticut	Presenter
2003	UF FAS-Fishing For Success	Assist the public as they learn fishing ethics
2003	National Shellfish Association	Audiovisual support
1999-03	R/V Discovery	Benthic sampling
2002	Oceans Day at Florida's Capital	University of Florida Co-representative
2001-03	University of Florida Dept. Fisheries and Aquatic Sciences Graduate Student Symposium	Presenter
2001-02	R/V Bellows cruises	Otter trawls, plankton tows & dredging
2000	North American Lake Management Society Symposium- Miami, Florida	Audiovisual support



# OMRI Listed®

The following product is OMRI Listed. It may be used in certified organic production or food processing and handling according to the USDA National Organic Program regulations.

## Product

BlueGreen Water Technologies Lake Guard Oxy Algaecide/Cyanobactericide Small Granules

## Company

BlueGreen US Water Technologies Inc.  
Bluegreen US Water Technologies  
2100 West Loop South, Suite 1100  
Houston TX 77027 United States

Status	Category	Issue date
Allowed with Restrictions	NOP: Sodium Carbonate Peroxyhydrate	18-Jul-2022
Product number	Class	Expiration date
bgu-17446	Crop Pest, Weed, and Disease Control	1-Sep-2023

## Restrictions

Federal law restricts the use of this substance in food crop production to approved food uses identified on the product label.

For use as an algaecide.

May only be used if the requirements of 205.206(e) are met, which requires the use of preventive, mechanical, physical, and other pest, weed, and disease management practices.

Executive Director/CEO

Product review is conducted according to the policies in the current *OMRI Policy Manual*® and based on the standards in the current *OMRI Standards Manual*®. To verify the current status of this or any OMRI Listed product, view the most current version of the *OMRI Products List*® at [OMRI.org](http://OMRI.org). OMRI listing is not equivalent to organic certification and is not a product endorsement. It cannot be construed as such. Final decisions on the acceptability of a product for use in a certified organic system are the responsibility of a USDA accredited certification agent. It is the operator's responsibility to properly use the product, including following any restrictions.



Organic Materials Review Institute  
P.O. Box 11558, Eugene, OR 97440-3758, USA  
541.343.7600 · [info@omri.org](mailto:info@omri.org) · [OMRI.org](http://OMRI.org)

## 1. PRODUCT & COMPANY IDENTIFICATION

1.1	Product Name:	<b>BLUEGREEN® LAKE GUARD™ OXY</b>
1.2	Chemical Name:	Sodium Percarbonate Mixture
1.3	Synonyms:	NA
1.4	Trade Names:	BlueGreen® Lake Guard™ Oxy
1.5	Product Use:	Algaecide/Biocide
1.6	Distributor's Name:	BlueGreen Water Technologies Ltd.
1.7	Distributor's Address:	3/15 Kachal Street, Tzur Hadassah, 9987500, Israel
1.8	Emergency Phone:	<b>CHEMTEL +1 (800) 255-3924</b>
1.9	Business Phone / Fax / Email:	Tel: +972 (2) 630-1166 / Fax: +972 (2) 630-1166 / Email: info@bgtechs.com

## 2. HAZARDS IDENTIFICATION

2.1	Hazard Identification:	Prepared in accordance with UN Globally Harmonized standards. Intended to comply with OSHA 29 CFR 1910.1200. Canadian WHMIS and Australian Work Health and Safety. <b>DANGER! MAY INTENSIFY FIRE – OXIDIZER. HARMFUL IF SWALLOWED. CAUSES SERIOUS EYE DAMAGE.</b> Classification: Ox. Sol. 2, Acute Tox. (oral) 4, Eye Dam. 1
2.2	Label Elements:	<p>Signal Word: DANGER</p> <p><b>Hazard Statements (H):</b> H272 – May intensify fire – oxidizer. H302 – Harmful if swallowed. H318 – Causes serious eye damage.</p> <p><b>Precautionary Statements (P):</b> P210 – Keep away from heat. P220 – Keep/Store away from clothing/ combustible materials. P221 – Take any precaution to avoid mixing with combustibles. P264 – Wash skin thoroughly after handling. P270 – Do not eat, drink, or smoke when using this product. P280 - Wear protective gloves/ protective clothing/ eye protection/ face protection. P301 + P312 – IF SWALLOWED: Call a POISON CENTER/ doctor if you feel unwell. P305 + P351 + P338 – IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. P310 – Immediately call a POISON CENTER/ doctor. P330 – Rinse mouth. P370 + P378 – In case of fire: Use dry sand, dry chemical, or alcohol-resistant foam for extinction. P391 – Collect spillage. P501 – Dispose of contents/ container to an approved waste disposal plant.</p>
2.3	Other Warnings:	<b>KEEP OUT OF REACH OF CHILDREN.</b>



## 3. COMPOSITION & INGREDIENT INFORMATION

CHEMICAL NAME(S)	CAS No.	RTECS No.	EINECS No.	%	EXPOSURE LIMITS IN AIR (mg/m <sup>3</sup> )									OTHER
					ACGIH		NOHSC			OSHA				
					TLV	STEL	ppm	ES-TWA	ES-STEL	ES-PEAK	PEL	STEL	IDLH	
SODIUM PERCARBONATE	15630-89-4	FG0750000	239-707-6	98	NA	NA	NF	NF	NF	NA	NA	NA		
PROPRIETARY INGREDIENTS	NA	NA	NA	2	NA	NA	NF	NF	NF	NA	NA	NA		


## 4. FIRST AID MEASURES

4.1	First Aid:	<p><b>Ingestion:</b> If ingested, do NOT induce vomiting. Rinse mouth with water. Never give anything by mouth to an unconscious person. Contact the nearest Poison Control Center or local emergency number. Provide an estimate of the time at which the material was ingested and the amount of the substance that was swallowed.</p> <p><b>Eyes:</b> Splashes are not likely; however, if product gets in the eyes, flush with copious amounts of lukewarm water for at least 15 minutes lifting upper and lower lids, occasionally. Get medical attention immediately.</p> <p><b>Skin:</b> IF ON SKIN: Wash with plenty of soap and water. Wash contaminated clothing before reuse. Wash off immediately with soap and plenty of water for at least 15 minutes. If symptoms persist, call a physician.</p> <p><b>Inhalation:</b> Remove victim to fresh air at once. If breathing difficult, administer oxygen. If breathing stops give artificial respiration. Keep person warm, quiet and get medical attention.</p>
4.2	Effects of Exposure:	<p><b>Ingestion:</b> If product is swallowed, may cause nausea, vomiting and/or diarrhea.</p> <p><b>Eyes:</b> Severely irritating to the eyes. Symptoms of overexposure may include redness, itching, irritation, and watering.</p> <p><b>Skin:</b> May be irritating to skin. Symptoms of overexposure may include redness, itching, irritation or burning sensation.</p> <p><b>Inhalation:</b> Persons accidentally exposed to dust, particularly during loading and unloading, may experience some effects including cough, wheezing and upper respiratory tract irritation.</p>

## 4. FIRST AID MEASURES – cont'd

4.3	Symptoms of Overexposure:	<p><b>Ingestion:</b> Severe irritation, nausea, abdominal pain, vomiting and/or diarrhea.</p> <p><b>Eyes:</b> Overexposure in eyes may cause redness, itching, swelling and watering.</p> <p><b>Skin:</b> Symptoms of skin overexposure may include redness, itching, and irritation of affected areas.</p> <p><b>Inhalation:</b> Coughing, irritation of mucous membranes and upper respiratory tract.</p>											
4.4	Acute Health Effects:	Severe irritation to eyes. Additionally, high concentrations of dusts can cause dizziness, headaches, and nausea.											
4.5	Chronic Health Effects:	Overexposure may trigger asthma-like symptoms in some sensitive individuals. May also induce skin sensitization and respiratory hypersensitivity.											
4.6	Target Organs:	Eyes, Respiratory System											
4.7	Medical Conditions Aggravated by Exposure:	Pre-existing disorders of the target organs (eyes, lungs).											
			<table border="1" style="width: 100%; border-collapse: collapse;"> <tr style="background-color: #0056b3; color: white;"> <td style="padding: 2px;">HEALTH</td> <td style="text-align: right; padding: 2px;">3</td> </tr> <tr style="background-color: #d9534f; color: white;"> <td style="padding: 2px;">FLAMMABILITY</td> <td style="text-align: right; padding: 2px;">0</td> </tr> <tr style="background-color: #e67e22; color: white;"> <td style="padding: 2px;">PHYSICAL HAZARDS</td> <td style="text-align: right; padding: 2px;">1</td> </tr> <tr style="background-color: #34495e; color: white;"> <td style="padding: 2px;">PROTECTIVE EQUIPMENT</td> <td style="text-align: right; padding: 2px;">A</td> </tr> <tr> <td style="padding: 2px;">EYES</td> <td></td> </tr> </table>	HEALTH	3	FLAMMABILITY	0	PHYSICAL HAZARDS	1	PROTECTIVE EQUIPMENT	A	EYES	
HEALTH	3												
FLAMMABILITY	0												
PHYSICAL HAZARDS	1												
PROTECTIVE EQUIPMENT	A												
EYES													

## 5. FIREFIGHTING MEASURES

5.1	Fire & Explosion Hazards:	<p>Oxidizing. Oxygen released in thermal decomposition may support combustion or accelerate burning when involved in a fire. Contact with combustible material may cause fire. Contact with flammables may cause fire or explosions. May ignite combustibles (wood, paper, oil, clothing, etc.). Risk of explosion if heated under confinement. May decompose explosively when heated or involved in a fire. During a fire, irritating and toxic gases may be generated by thermal decomposition or combustion. Hazardous combustion products include oxides of carbon (CO, CO<sub>2</sub>) and oxygen. Runoff may create fire or explosion hazard.</p>	
5.2	Extinguishing Methods:	Use water. Do not use dry chemicals or foams. CO <sub>2</sub> or Halon may provide limited control. Flood fire area with water from a distance. Move containers from fire area if you can do it without risk. Cool containers with flooding quantities of water until well after fire is out.	
5.3	Firefighting Procedures:	Firefighters should wear self-contained breathing apparatus and full firefighting turnout gear. Use personal protection equipment. Do not move cargo or vehicle if cargo has been exposed to heat. oxidizer. May ignite combustibles (wood, paper, oil, clothing, etc.). Move containers from fire area if you can do it without risk. Fight fire from maximum distance or use unmanned hose holders or monitor nozzles. ALWAYS stay away from tanks engulfed in fire. For massive fire, use unmanned hose holders or monitor nozzles; if this is impossible withdraw from area and let fire burn.	



## 6. ACCIDENTAL RELEASE MEASURES

6.1	Spills:	<p>Ensure adequate ventilation. Avoid contact with eyes or clothing. Evacuate personnel to safe areas. Keep people away from and upwind of spill/leak. ELIMINATE all ignition sources (no smoking, flares, sparks, or flames in immediate area). Do not touch damaged containers or spilled material unless wearing appropriate protective clothing. See section 8 for more information. Stop leak if you can do it without risk. Use personal protective equipment as required. Keep combustibles (wood, paper, oil, etc.) away from spilled material. DO NOT GET WATER INSIDE CONTAINERS. Ventilate the area. Refer to protective measures listed in Sections 7 and 8.</p> <p>Stop leak if you can do it without risk. Cover with DRY earth, DRY sand or other non-combustible material followed with plastic sheet to minimize spreading or contact with rain.</p> <p>With clean shovel place material into clean, dry container and cover loosely; move containers from spill area. Flush area with flooding quantities of water. Prevent product from entering drains. Cover powder spill with plastic sheet or tarp to minimize spreading and keep powder dry.</p>
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## 7. HANDLING & STORAGE INFORMATION

7.1	Work & Hygiene Practices:	<p>Use personal protection equipment. Avoid contact with eyes, or clothing. Keep away from heat, hot surfaces, sparks, open flames, and other ignition sources. No smoking. Do not eat, drink, or smoke when using this product. Remove contaminated clothing and shoes. Use with local exhaust ventilation. Handle in accordance with good industrial hygiene and safety practice. Avoid formation of dust and aerosols. Avoid breathing dusts from this product. Avoid excessive dust generation. Further, processing of solid materials may result in the formation of combustible dusts. The potential for combustible dust formation should be taken into consideration before additional processing occurs. Provide appropriate exhaust ventilation at places where dust is formed. Use in a well-ventilated location (e.g., local exhaust ventilation, fans). After use, wash hands and exposed skin with soap and water. Do not eat, drink, or smoke while handling product.</p>
7.2	Storage & Handling:	Keep container tightly closed in a dry and well-ventilated place. Keep this material away from heat, sparks, and open flame. Open containers slowly on a stable surface. Store containers in a cool, dry location, away from direct sunlight, other light sources, or sources of intense heat. Store away from incompatible materials (See Section 10).
7.3	Special Precautions:	Storage class (TRGS 510): 5.1B: Oxidizing Materials

## 8. EXPOSURE CONTROLS & PERSONAL PROTECTION

8.1	Exposure Limits: ppm (mg/m <sup>3</sup> )	ACGIH		NOHSC			OSHA		OTHER	
		TLV	STEL	ES-TWA	ES-STEL	ES-PEAK	PEL	STEL		IDLH
		CHEMICAL NAME(S)								
		SODIUM PERCARBONATE		NA	NA	NF	NF	NF	NA	NA
8.2	Ventilation & Engineering Controls:	Use local or general exhaust ventilation to effectively remove and prevent buildup of dust or vapors generated from the handling of this product. Ensure appropriate decontamination equipment is available (e.g., sink, safety shower, eye-wash station). Periodic medical exams of exposed workers may be required.								
8.3	Respiratory Protection:	No protective equipment is needed under normal use conditions. If exposure limits are exceeded or irritation is experienced, ventilation and evacuation may be required. Keep the exposure within legal limits. In the worker's breathing zone and the general area, dusts must be kept below the TLVs, and the equivalent exposure must compute to less than one. Keep exposure as low as possible. When dusts are generated, and respiratory protection is needed, use only protection authorized by 29 CFR §1910.134, applicable U.S. State regulations, or the Canadian CAS Standard Z94.4-93 and applicable standards of Canadian Provinces, EC member States, or Australia.								
8.4	Eye Protection:	Always use protective eyewear (e.g., tightly sealing chemical safety goggles) when handling, or when cleaning spills or leaks. Contact lenses pose a special hazard; soft lenses may absorb and concentrate irritants. If necessary, refer to U.S. OSHA 29 CFR §1910.133, Canadian standards, or the European Standard EN166. 								
8.5	Hand Protection:	Wear suitable, impervious gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands. 								
8.6	Body Protection:	Wear suitable protective clothing. Long sleeved clothing. Chemical resistant apron. Wear fire/flame resistant/retardant clothing. If necessary, refer to appropriate standards of Canada, the E.C. member states, or U.S. OSHA.								

## 9. PHYSICAL & CHEMICAL PROPERTIES

9.1	Appearance:	Solid, white powder
9.2	Odor:	Odorless
9.3	Odor Threshold:	NA
9.4	pH:	10.5
9.5	Melting Point/Freezing Point:	Decomposition at T>140°C
9.6	Initial Boiling Point/Boiling Range:	NA
9.7	Flashpoint:	>230°C
9.8	Upper/Lower Flammability Limits:	NA
9.9	Vapor Pressure:	NA
9.10	Vapor Density:	(25 °C) negligible
9.11	Relative Density:	1.93 g/cm <sup>3</sup>
9.12	Solubility:	Soluble in water, solubility 140 g/L
9.13	Partition Coefficient (log P <sub>ow</sub> ):	NA
9.14	Autoignition Temperature:	NA
9.15	Decomposition Temperature:	NA
9.16	Viscosity:	NA
9.17	Other Information:	Bulk density: 700-1200 kg/m <sup>3</sup>

## 10. STABILITY & REACTIVITY

10.1	Stability:	May cause fire or explosion, strong oxidizer.
10.2	Hazardous Decomposition Products:	Oxides of carbon, oxygen.
10.3	Hazardous Polymerization:	Spontaneous polymerization can occur.
10.4	Conditions to Avoid:	Exposure or contact to extreme temperatures, heat, sparks, flames, incompatible chemicals, moisture.
10.5	Incompatible Substances:	Organic material. Combustible material. Hydrocarbons. Strong acids. Strong bases. Strong oxidizing agents.

Prepared to OSHA, ACC, ANSI, NOHSC, WHMIS, GHS & EU Standards      SDS Revision: 1.5      SDS Revision Date: 12/22/2021

## 11. TOXICOLOGICAL INFORMATION

11.1	Routes of Entry:	Inhalation: YES	Absorption: YES	Ingestion: YES	
11.2	Toxicity Data:	LD <sub>50</sub> (oral, mice): >5009.1 mg/kg. Causes serious eye irritation (OECD Test Guideline 405).			
11.3	Acute Toxicity:	Serious irritation to eyes.			
11.4	Chronic Toxicity:	This material may aggravate any pre-existing skin condition (e.g., dermatitis).			
11.5	Suspected Carcinogen:	No			
11.6	Reproductive Toxicity:	This product is not reported to produce reproductive toxicity in humans.			
	Mutagenicity:	This product is not reported to produce mutagenic effects in humans.			
	Embryotoxicity:	This product is not reported to produce embryotoxic effects in humans.			
	Teratogenicity:	This product is not reported to cause teratogenic effects in humans.			
11.7	Irritancy of Product:	<b>General Nuisance Dusts:</b> Nuisance dusts, which are essentially nontoxic and chemically non-irritating. Skin contact has shown no problems other than possible drying and mechanical irritation. Eye contact can produce particulate irritation. Excessive inhalation can produce mild pulmonary irritation and possible non-disabling slight fibrosis of the lungs. The product can cause allergic skin reactions (e.g., rashes, welts, dermatitis) upon prolonged or repeated exposure.			
11.8	Biological Exposure Indices:	NE			
11.9	Physician Recommendations:	Treat symptomatically.			

## 12. ECOLOGICAL INFORMATION


12.1	Environmental Stability:	There are no specific data available for this product.
12.2	Effects on Plants & Animals:	There are no specific data available for this product.
12.3	Effects on Aquatic Life:	LC <sub>50</sub> (Zebra fish, 96h): > 100.0 mg/L. WGK: 1

## 13. DISPOSAL CONSIDERATIONS


13.1	Waste Disposal:	Disposal should be in accordance with local, state, and federal regulations. Dispose of in accordance with federal, state, and local regulations.
13.2	Special Considerations:	Offer surplus and non-recyclable solutions to a licensed disposal company. Contact a licensed professional waste disposal service to dispose of this material.

## 14. TRANSPORTATION INFORMATION

The basic description (ID Number, proper shipping name, hazard class & division, packing group) is shown for each mode of transportation. Additional descriptive information may be required by 49 CFR, IATA/ICAO, IMDG and the CTDGR.

14.1	49 CFR (GND):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 5.0 kg)	
14.2	IATA (AIR):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 1.0 kg)	
14.3	IMDG (OCN):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 5.0 kg)	
14.4	TDGR (Canadian GND):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 5.0 kg)	
14.5	ADR/RID (EU):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 5.0 kg)	
14.6	SCT (MEXICO):	UN3378, CARBONATO DE SODIO PEROXIHDRATADO, 5.1, III, CANT. LTDA. (IP VOL ≤ 5.0 kg)	
14.7	ADGR (AUS):	UN3378, SODIUM CARBONATE PEROXYHYDRATE, 5.1, III, LTD QTY (IP VOL ≤ 5.0 kg)	
14.8	U.S. CENSUS/FOREIGN TRADE	SCHEDULE B: 2836.99.0000 or 2836.99.0007	

## 15. REGULATORY INFORMATION

15.1	SARA Reporting Requirements:	This product does not contain any substances subject to SARA Title III, Section 313 reporting requirements.	
15.2	SARA TPC:	There are no specific Threshold Planning Quantities for the components of this product.	
15.3	TSCA Inventory Status:	The components of this product are listed on the TSCA Inventory.	
15.4	CERCLA Reportable Quantity:	NA	
15.5	Other Federal Requirements:	NA	
15.6	Other Canadian Regulations:	This product has been classified according to the hazard criteria of the Controlled Products Regulations (CPR) and the SDS contains all of the information required by the CPR. The components of this product are listed on the DSL/NDL. None of the components of this product are listed on the Priorities Substances List. WHMIS C, D2B (Oxidizing, Other Harmful Effects)	
15.7	State Regulatory Information:	No ingredients in this product, present in a concentration of 1.0% or greater, are listed on any of the following state criteria lists: California Proposition 65 (CA65), Delaware Air Quality Management List (DE), Florida Toxic Substances List (FL), Massachusetts Hazardous Substances List (MA), Michigan Critical Substances List (MI), Minnesota Hazardous Substances List (MN), New Jersey Right-to-Know List (NJ), New York Hazardous Substances List (NY), Pennsylvania Right-to-Know List (PA), Washington Permissible Exposures List (WA), Wisconsin Hazardous Substances List (WI).	
15.8	Other Requirements:	NA	



## 16. OTHER INFORMATION

16.1	Other Information:	<p><b>DANGER! MAY INTENSIFY FIRE – OXIDIZER. HARMFUL IF SWALLOWED. CAUSES SERIOUS EYE DAMAGE.</b>            Keep away from heat. Keep/Store away from clothing/combustible materials. Take any precaution to avoid mixing with combustibles. Wash skin thoroughly after handling. Do not eat, drink, or smoke when using this product. Wear protective gloves/ protective clothing/ eye protection/ face protection. IF SWALLOWED: Call a POISON CENTER/ doctor if you feel unwell. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/doctor. Rinse mouth. In case of fire: Use dry sand, dry chemical, or alcohol-resistant foam for extinction. Collect spillage.  <b>KEEP OUT OF REACH OF CHILDREN.</b></p>	
16.2	Terms & Definitions:	See last page of this Safety Data Sheet.	
16.3	Disclaimer:	<p>This Safety Data Sheet is offered pursuant to OSHA's Hazard Communication Standard, 29 CFR §1910.1200. Other government regulations must be reviewed for applicability to this product. To the best of ShipMate's &amp; BlueGreen Water Technologies Ltd.'s knowledge, the information contained herein is reliable and accurate as of this date; however, accuracy, suitability or completeness is not guaranteed and no warranties of any type, either expressed or implied, are provided. The information contained herein relates only to the specific product(s). If this product(s) is combined with other materials, all component properties must be considered. Data may be changed from time to time. Be sure to consult the latest edition.</p>	
16.4	Prepared for:	<p><b>BlueGreen Water Technologies Ltd.</b>            3/15 Kachal Street            Tzur Hadassah, 9987500, Israel            Tel: +972 (2) 630-1166            Fax: +972 (2) 630-1166            Email: info@bgtechs.com  <a href="https://bgtechs.com/">https://bgtechs.com/</a></p>	
16.5	Prepared by:	<p><b>ShipMate, Inc.</b>            P.O. Box 787            Sisters, Oregon 97759-0787 USA            Tel: +1 (310) 370-3600            Fax: +1 (310) 370-5700  <a href="https://shipmate.com/">https://shipmate.com/</a></p>	

## DEFINITION OF TERMS

A large number of abbreviations and acronyms appear on a SDS. Some of these that are commonly used include the following:

### GENERAL INFORMATION:

<b>CAS No.</b>	Chemical Abstract Service Number
<b>RTECS No.</b>	Registry of Toxic Effects of Chemical Substances Number
<b>EINECS No.</b>	European Inventory of Existing Commercial Chemical Substances Number

### EXPOSURE LIMITS IN AIR:

<b>ACGIH</b>	American Conference on Governmental Industrial Hygienists
<b>IDLH</b>	Immediately Dangerous to Life and Health
<b>NOHSC</b>	National Occupational Health and Safety Commission (Australia)
<b>OSHA</b>	U.S. Occupational Safety and Health Administration
<b>PEL</b>	Permissible Exposure Limit
<b>STEL</b>	Short Term Exposure Limit
<b>TLV</b>	Threshold Limit Value
<b>TWA</b>	Time Weighted Average

### FIRST AID MEASURES:

<b>CPR</b>	Cardiopulmonary resuscitation - method in which a person whose heart has stopped receives manual chest compressions and breathing to circulate blood and provide oxygen to the body.
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### HAZARDOUS MATERIALS IDENTIFICATION SYSTEM: HMIS

#### HEALTH, FLAMMABILITY & REACTIVITY RATINGS:

0	Minimal Hazard
1	Slight Hazard
2	Moderate Hazard
3	Severe Hazard
4	Extreme Hazard

HEALTH
FLAMMABILITY
PHYSICAL HAZARDS
PERSONAL PROTECTION

#### PERSONAL PROTECTION RATINGS:

<b>A</b>	
<b>B</b>	
<b>C</b>	
<b>D</b>	
<b>E</b>	
<b>F</b>	

<b>G</b>	
<b>H</b>	
<b>I</b>	
<b>J</b>	
<b>K</b>	
<b>X</b>	Consult your supervisor or SOPs for special handling directions.

Safety Glasses	Splash Goggles	Face Shield & Protective Eyewear	Gloves
Boots	Protective Apron	Protective Clothing & Full Suit	Dust Respirator
Full Face Respirator	Dust & Vapor Half-Mask Respirator	Full Face Respirator	Airline Hood/Mask or SCBA

#### OTHER STANDARD ABBREVIATIONS:

<b>Carc</b>	Carcinogenic
<b>Irrit</b>	Irritant
<b>NA</b>	Not Available
<b>NR</b>	No Results
<b>ND</b>	Not Determined
<b>NE</b>	Not Established
<b>NF</b>	Not Found
<b>SCBA</b>	Self-Contained Breathing Apparatus
<b>Sens</b>	Sensitization
<b>STOT RE</b>	Specific Target Organ Toxicity – Repeat Exposure
<b>STOT SE</b>	Specific Target Organ Toxicity – Single Exposure

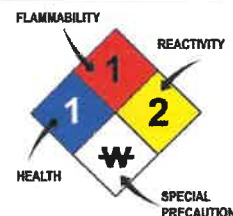
### NATIONAL FIRE PROTECTION ASSOCIATION: NFPA

#### FLAMMABILITY LIMITS IN AIR:

<b>Autoignition Temperature</b>	Minimum temperature required to initiate combustion in air with no other source of ignition
<b>LEL</b>	Lower Explosive Limit - lowest percent of vapor in air, by volume, that will explode or ignite in the presence of an ignition source
<b>UEL</b>	Upper Explosive Limit - highest percent of vapor in air, by volume, that will explode or ignite in the presence of an ignition source

#### HAZARD RATINGS:

<b>0</b>	Minimal Hazard
<b>1</b>	Slight Hazard
<b>2</b>	Moderate Hazard
<b>3</b>	Severe Hazard
<b>4</b>	Extreme Hazard
<b>ACD</b>	Acidic
<b>ALK</b>	Alkaline
<b>COR</b>	Corrosive
<b>W</b>	Use No Water
<b>OX</b>	Oxidizer
<b>TREFOIL</b>	Radioactive



#### TOXICOLOGICAL INFORMATION:

<b>LD<sub>50</sub></b>	Lethal Dose (solids & liquids) which kills 50% of the exposed animals
<b>LC<sub>50</sub></b>	Lethal concentration (gases) which kills 50% of the exposed animal
<b>ppm</b>	Concentration expressed in parts of material per million parts
<b>TD<sub>01</sub></b>	Lowest dose to cause a symptom
<b>TCLO</b>	Lowest concentration to cause a symptom
<b>TD<sub>01</sub>, LD<sub>01</sub>, &amp; LD<sub>01</sub> or TC, TC<sub>01</sub>, LC<sub>01</sub>, &amp; LC<sub>01</sub></b>	Lowest dose (or concentration) to cause lethal or toxic effects
<b>IARC</b>	International Agency for Research on Cancer
<b>NTP</b>	National Toxicology Program
<b>RTECS</b>	Registry of Toxic Effects of Chemical Substances
<b>BCF</b>	Bioconcentration Factor
<b>TL<sub>m</sub></b>	Median threshold limit
<b>log K<sub>ow</sub> or log K<sub>oc</sub></b>	Coefficient of Oil/Water Distribution

#### REGULATORY INFORMATION:

<b>WHMIS</b>	Canadian Workplace Hazardous Material Information System
<b>DOT</b>	U.S. Department of Transportation
<b>TC</b>	Transport Canada
<b>EPA</b>	U.S. Environmental Protection Agency
<b>DSL</b>	Canadian Domestic Substance List
<b>NDSL</b>	Canadian Non-Domestic Substance List
<b>PSL</b>	Canadian Priority Substances List
<b>TSCA</b>	U.S. Toxic Substance Control Act
<b>EU</b>	European Union (European Union Directive 67/548/EEC)
<b>WGK</b>	Wassergefährdungsklassen (German Water Hazard Class)
<b>TRGS900</b>	Die Technischen Regeln für Gefahrstoffe (TRGS) - The Technical Rules for Hazardous Substances (TRGS) - Germany

#### WORKPLACE HAZARDOUS MATERIALS IDENTIFICATION (WHMIS) SYSTEM:

Class A	Class B	Class C	Class D1	Class D2	Class D3	Class E	Class F
Compressed	Flammable	Oxidizing	Toxic	Irritation	Infectious	Corrosive	Reactive

#### CLP/GHS (1272/2008/EC) PICTOGRAMS:

GHS01	GHS02	GHS03	GHS04	GHS05	GHS06	GHS07	GHS08	GHS09
Explosive	Flammable	Oxidizer	Pressurized	Corrosive	Toxic	Harmful/Irritating	Health Hazard	Environment