Cyanotoxin Monitoring in the Delta: Leveraging existing USGS and DWR field efforts to identify cyanotoxin occurrence, duration, and drivers

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November 16, 2021

Problem Statement

One major impediment to improved understanding and prediction of harmful algal blooms (HABs) and the cyanotoxins they produce is the dearth of systematic collection of observational data across both space and time. HABs, which in freshwater comprise mostly cyanobacteria (cyanoHABs), are distributed worldwide and are a growing concern because they can adversely affect drinking water supplies, interfere with water transfers, harm aquatic organisms, and potentially harm humans and wildlife. Worldwide, the distribution and abundance of cyanoHABs are intensified by increased nutrient loads from agriculture and urban runoff, atmospheric deposition, global warming, and droughts. It is most often the cyanotoxins produced by these organisms that are the hazard rather than the organisms themselves – which may or may not produce toxins – so improved monitoring efforts seek to combine cyanobacterial detection with measurement of the toxins themselves.

Identifying drivers of cyanoHABs and their associated toxins requires an understanding of the conditions that foster their growth as well as hydrologic drivers that then transport them through the ecosystem. Environmental factors that have been attributed to the occurrence of cyanoHABs and the toxins they produce include nutrient concentrations, light conditions, water temperature, hydrologic conditions, water residence time, and meteorological conditions. These factors change rapidly in aquatic systems, particularly in hydrologically complex and tidal estuaries like the Delta (Kraus et al., 2017). Thus, a robust monitoring program for cyanoHABs and cyanotoxins requires investing in collection of a wide array of parameters, a task that is often cost prohibitive. Due to the high costs of these efforts, there has been limited and sporadic cyanotoxin sampling in the Delta to date (Lehman et al. 2005, 2008, 2017; Otten et al. 2017). However, we do know from this work that cyanoHABs occur each year and negatively impact aquatic species at multiple trophic levels in the estuary (Lehman et al. 2010, 2017). Here we propose to add cyanotoxin sampling to existing water quality monitoring programs run by the US Geological Survey California Water Science Center (USGS) and California Department of Water Resources (DWR) that already collect flow, water quality, nutrient, and phytoplankton data (Table 1).

Another challenge for monitoring cyanotoxins is that the occurrence of these compounds can be ephemeral and/or episodic. Thus, discrete sampling programs that occur on a monthly or even bimonthly interval can miss key events and underestimate cyanotoxin risk, or if they capture a high-concentration event can give a false impression that cyanotoxins are a widespread health hazard. The use of SPATTs

(Solid Phase Adsorption Toxin Tracking) samplers helps address this issue by providing a temporally integrated signal of dissolved cyanotoxin concentrations (Kudela, 2017; Howard et al, 2017; Peacock et al., 2018, Howard et al., 2018). SPATT samplers have been used as a compliment to traditional monitoring programs and can elucidate toxin dynamics and environmental drivers. SPATT samplers have detected HAB toxins when simultaneous "grab" samples of water have failed to detect the same toxins in a given waterway as SPATT captures ephemeral events that may be missed by whole water sampling, including the prevalence of toxins, and exhibits more sensitivity compared with grab samples (Lane et al., 2010, Kudela, 2011; Howard et al., 2017; Kudela, 2017; Peacock et al., 2018). A timeseries of water (particulate fraction) and SPATT samples were collected in San Francisco Bay (SFB) from 2011 to 2016 and analyzed for both cyanotoxins and marine toxins (Peacock et al., 2018). The SPATT results indicated ubiquitous toxins throughout SFB, however, the particulate water samples only captured toxins during some timepoints and generally indicated toxins were not very prevalent. Both particulate and dissolved toxins are concentrated by shellfish (Miller et al., 2010; Gibble et al., 2016) and additional studies indicated multiple toxins were routinely present in mussels indicating a potential for transfer of toxins throughout the food web (Gibble et al., 2016; Peacock et al., 2018). Therefore, using SPATT as a monitoring tool provided insight into the toxin detections in mussel samples, and the potential for transfer to the food web that the grab samples did not capture (Peacock et al., 2018).

Table 1. Configuration of USGS and DWR continuous monitoring stations.

Type	Description
ADCP, Pressure Sensors	Flow, Discharge, Gauge Height
Infrastructure	Data Collection Platform (Enclosure, Datalogger, wire and cable, telemetry, solar panels, regulators and batteries)
YSI EXO	Temp/Cond sensor
	pH sensor
	D.O. sensor
	Turbidity sensor
	fDOM sensor*
	Total algae sensor (Total chlorophyll (fCHL) and Phycocyanin (PC)
	Central Wiper
	signal output adaptors
SUNA Nitrate Analyzer*	SUNA Nitrate Analyzer*
bbe Fluoroprobe**	chlorophyll attributed to four phytoplankton classes (cyanobacteria, diatoms, green algae, chlorophytes)

*USGS stations only; **planned for MDM, JPT, DEC, CFL stations

Background

The Sacramento-San Joaquin Delta (Delta) serves as critical aquatic habitat and as a vital drinking water resource for almost 30 million Californians. It is also a physically, biologically, and hydrologically complex system, receiving flows from the Sacramento and San Joaquin Rivers, which drain approximately 40% of California and then move through and merge within the Delta, a maze-like network of interconnected channels and sloughs (Figure 1). Analysis of long-term observational data demonstrate that the Delta is in a state of severe ecological decline (Sommer et al. 2007; Thomson et al. 2010). In particular, the structure and function of habitats and the lower trophic levels has been transformed

through, invasive aquatic macrophytes, localized issues with low dissolved oxygen, excessive anthropogenic nutrients, and cyanoHABs.

Information about cyanoHABs and cyanotoxins in the Delta are available for the summer and fall months (Lehman et al. 2005, 2008, 2010, 2017; Otten et al. 2017). However, with warmer conditions due to climate change, blooms are starting earlier and lasting longer, suggesting that more extensive temporal sampling is needed to determine the current bloom impact (Lehman et al. 2017). The spatial extent of cyanoHABs is also changing; while these organisms have been detected in the Central and Southern Delta for many years, they have more recently been observed in the northern Delta including the Cache Slough Complex (Figure 1).

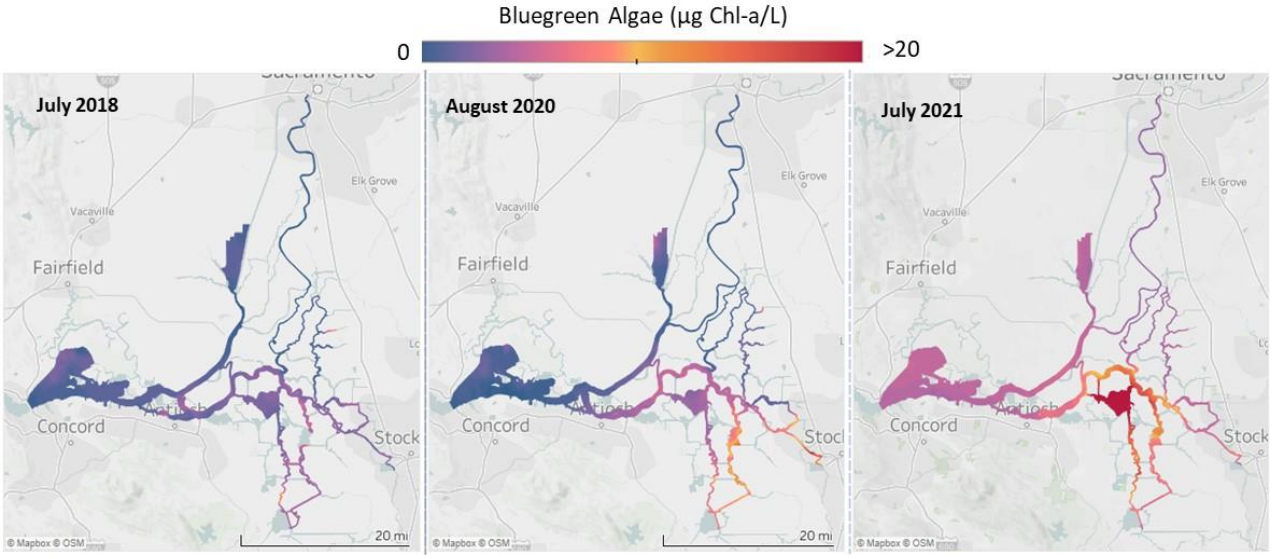

Figure 1. Data collected in July 2018, August 2020 and July 2021 during high resolution boat-based mapping surveys of the study area (Sacramento-San Joaquin Delta, California). Color gradient shows variation in the chlorophyll-a pool attributed to blue green algae (i.e.cyanobacteria) measured using a bbe Fluoroprobe (FP).

Previously, the USGS received internal funding to collect cyanotoxins at two USGS continuous monitoring stations in the Delta (Jersey Point (JPT) and Decker (DEC), Figure 2). Then in 2020 the Delta Regional Monitoring Program (DRMP) funded the collection of samples for cyanotoxin analyses at four additional stations: two run by the USGS and two run by DWR (Figure 2). With the internal USGS and DRMP funding in 2020-2021 USGS was able to monitor cyanotoxins in 6 sites, however, both these funding sources expire in early 2022. Fortunately, in 2021 the USGS received funding from the Delta Science Program (DSP) to continue cyanotoxin collection at 5 of these sites. This funding will begin in Spring 2022, but funding was not sufficient enough to cover all previous 6 sites. Without additional funding, cyanotoxins will have to be dropped from one of the monitoring stations. The USGS was also previously funded to collect cyanotoxins during the high-frequency mapping surveys (Figure 1), but this funding also expires in December 2021.

The USGS and DWR have also been coordinating with Dr. Hans Paerl who was funded by the DSP to study aerosolized cyanotoxins in the Delta in 2022. Dr. Paerl's team will be collecting samples at the

Stockton Waterfront and Discovery Bay to study the entrainment of cyanobacteria and cyanotoxins in the atmosphere. Scientists in the USGS, DWR, and the Paerl Team are collaborating to leverage the expertise and resources of these teams to help ensure successful cyanoHABs research projects in 2022.

In addition to cyanoHAB specific projects, the U.S. Geological Survey (USGS) California Water Science Center (CAWSC) and the California Department of Water Resources (CDWR) operate a network of continuous flow and water quality monitoring stations across the Delta (Figure 2). Stations are instrumented with multiparameter sondes that measure water temperature, specific conductance, turbidity, pH, dissolved oxygen (DO), fluorescence of "total" chlorophyll (fCHL), as well as a sensor that measures nitrate (Table 1). These stations are serviced approximately monthly, and at the same time interval discrete water samples are collected to validate and calibrate these instruments (e.g., chlorophyll-a, nitrate) as well as to collect samples for laboratory analyses (e.g., phosphorus, ammonium, dissolved organic nitrogen, phytoplankton identification and enumeration) (Table 2). Most stations report flow, water velocity, and stage, allowing for calculation of constituent fluxes.

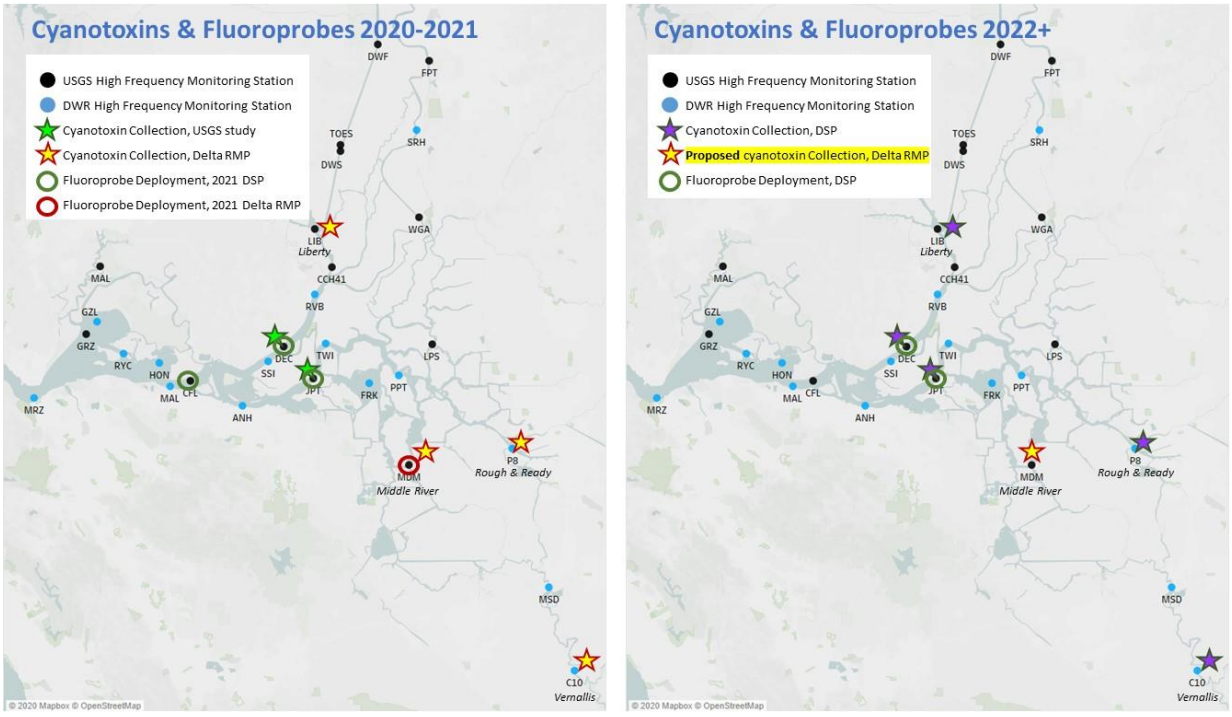

Figure 2. Map of the Delta showing locations of USGS (black circles) and DWR (blue circles) continuous monitoring stations. LEFT panel shows cyanotoxin and fluoroprobe monitoring in 2020-2021 funded by Delta RMP, Delta Science Program (DSP), and internal USGS funds. Funding for all these projects ends in early 2022. RIGHT panel shows cyanotoxin and fluoroprobe monitoring funded by DSP beginning in 2022. The yellow star in the right panel shows the MDM location for cyanotoxin monitoring proposed in this study.

Study Objectives

To provide a more comprehensive picture of the seasonal variation of HABs and their associated toxins in the Delta, this study would:

Task 1: Collect a full year of measurements of cyanotoxins at one station (Middle River, MDM) in the Delta that already have existing, robust monitoring programs, to supplement DSP funding and maintain a network of 6 cyanotoxin monitoring stations in the Delta.

Task 2: Monitor cyanoHAB bloom formation in the Stockton Waterfront by collecting vertical profiles of cyanobacteria to understand how bloom density, distribution, and toxins change over summer 2022.

Task 3: Collect cyanotoxins during USGS high-resolution mapping surveys in 2022.

Relevance to RMP Management Questions

The data gathered will provide important information to help stakeholders engaged in the Delta Nutrient Research Plan to determine whether nutrient concentrations and future management of nutrient concentrations could affect the initiation, duration, and source of cyanobacterial species and toxins in the Delta. Simultaneous collection of nutrients, phytoplankton and cyanotoxin information along with other water quality parameters (temperature, specific conductance, DO, pH) also will allow researchers to investigate how the suite of conditions along with nutrient concentrations contribute to HABs. The objectives of the project and how the information will be used relative to the RMP's high-level management questions are summarized in Table 2.

Table 2. Study objectives and questions relevant to RMP management questions.

Core Management Question	Study Objectives/Questions
Status & Trends Is there a problem or are there signs of a problem? a. Is water quality currently, or trending towards, adversely affecting beneficial uses of the Delta? b. Which constituents may be impairing beneficial uses in subregions of the Delta? c. Are trends similar or different across different subregions of the Delta?	**How do harmful algal blooms and cyanotoxin concentrations vary spatially and temporally year-round?** **How are ambient concentrations and trends in HABs and cyanotoxins affected by variability in water quality conditions, particularly nutrients?** Collect cyanotoxin data and associated phytoplankton and water quality variables year-round from MDM for one year and collect discrete cyanotoxin samples during mapping surveys. Year-round surveys will enable a more comprehensive assessment of the variation of HABs and cyanotoxins and how they are impacted by water quality conditions, flow (i.e., drought) including nutrient concentration.
Sources, Pathways, Loadings, and Processes Which sources and processes are most important to understand and quantify? a. Which sources, pathways, loadings, and processes (e.g., transformations, bioaccumulation) contribute most to identified problems?	**Which areas of the Delta are cyanotoxins produced and how are they transported?** **Which sources and levels of nutrients are more closely linked to HAB and toxin formation?** Provide online access to data and spatial and temporal trend plots of nutrient concentrations, associated water quality conditions, phytoplankton abundance and cyanotoxins for managers and scientists.

Core Management Question	Study Objectives/Questions
b. What is the magnitude of each source and/or pathway (e.g., municipal wastewater, atmospheric deposition)? c. What are the magnitudes of internal sources and/or pathways (e.g. benthic flux) and sinks in the Delta?	Vertical profiles of toxins and nutrients at Stockton Waterfront.
Forecasting scenarios a. How do ambient water quality conditions respond to different management scenarios b. What constituent loads can the Delta assimilate without impairment of beneficial uses? c. What is the likelihood that the Delta will be water quality-impaired in the future?	**Are cyanotoxin concentrations linked with nutrient concentrations, forms and ratios?** **How will changes to nutrient inputs to the Delta (e.g., WWTP upgrades) affect the development of HABs and cyanotoxins?** Identifying current linkages between environmental drivers (nutrients, flow, temperature) on HAB formation, initiation, and duration will assist modeling and targeted data analyses.
Effectiveness Tracking a. Are water quality conditions improving as a result of management actions such that beneficial uses will be met? b. Are loadings changing as a result of management actions?	Data collected by this study can be used to determine whether cyanotoxins are at concentrations of concern in the Delta and will help managers develop future monitoring programs. Data collected by this study will help us understand where cyanotoxins are produced and how they are transported in the Delta.

Study Approach

Task 1: Cyanotoxin monitoring at Middle River

We will continue to measure cyanotoxins at the Middle River site (MDM). Cyanotoxins will be measured with discrete water samples and solid phase adsorption toxin tracking (SPATT) samplers. The MDM station is currently equipped with a YSI EXO (water temperature, specific conductance, turbidity, pH, dissolved oxygen, chlorophyll-a/BGA), a SUNA nitrate analyzer, and a bbe Fluoroprobe (Table 1).

Previous studies suggest that cyanotoxin concentrations in the Delta are higher in the summer and fall and lower in the winter and spring, thus we will collect samples approximately every 4 weeks (monthly) in the winter and spring, and approximately every 2 weeks in the summer and fall, for a total of 18 sample dates at MDM. Monthly (12 per year) water samples are collected at these stations under existing USGS and DWR programs, so additional samples for nutrients, phytoplankton enumeration, and picoplankton counts only are needed under this study for the 6 additional sampling dates (Table 3).

SPATT samples: The use of SPATT samplers (Figure 3) has recently been refined as a monitoring tool to compliment traditional discrete sampling programs by providing a time-integrated indicator of dissolved toxin presence (Lane et al., 2010; Kudela, 2011; Howard et al., 2017; Kudela, 2017, Peacock et al., 2018; Roue and others, 2018). SPATT samplers will be constructed in the USGS laboratory following methods described in Howard and others (2018). SPATTs will be deployed adjacent to sonde measurements. Each SPATT will be deployed for approximately two weeks; when one sampler is removed from the station a new one will immediately be deployed in its place. SPATT bags will be placed in ziplock bags, placed immediately on dry ice in the field, kept frozen (-80° C), and then sent to the laboratory (Lumigen Instrument Center) for extraction and analysis. All (100%) SPATTs will undergo analysis via the method of liquid chromatography with tandem mass

spectrometry (LCMS-MS) for the detection of cyanotoxins listed in Table 2. Upon review of LCMS-MS data – a subset of samples (~20%) will be selected for analysis via the method of enzyme-linked immunosorbent assay (ELISA) by BSA Environmental Services, which is limited to the detection of four cyanotoxins (Table 3). Cyanotoxin methods of analysis differ by state and federal entities – analyses of SPATTs from this study using both analytical methods allow for data and method comparability across different HABs-funded studies.

Figure 3. Photo showing the planned system for deploying SPATT at fixed locations.

Discrete water samples: In addition to collecting SPATTs, we will collect discrete whole water samples concurrent with the removal/placement of SPATTs (approx.18 times per year), which is concurrent with sample collection for analytes listed in Table 3. Whole water samples will be placed immediately on dry ice in the field, kept frozen (-80° C), and then sent to the laboratory (Lumigen Instrument Center) for analysis. All (100%) whole water samples will undergo analysis via LCMS-MS and – upon review of LCMS-MS data – a subset of samples (~20%) will be selected for analysis via ELISA (BSA Environmental Services). Again, analysis of discrete water samples from this study using both analytical methods allows for data and method comparability across different HABs-funded studies.

The goal of implementing SPATT into this proposed study is as a monitoring tool to provide a robust, comprehensive approach to determining toxin patterns and dynamics within the Delta that traditional water grab samples alone can miss. We are very much aware of all the confounding factors that make SPATT cyanotoxin collection challenging to interpret compared to whole water samples, particularly because relating cyanotoxin data obtained from SPATT samplers to a health advisory threshold is not straightforward. The study objective is not to relate SPATT results to human health regulations, but rather to use SPATT as a separate, complementary sampling tool with water grabs to elucidate the prevalence of toxins and to capture ephemeral events that water grab samples can miss. That is why we are collecting SPATT only in conjunction with the more traditional whole water method, which is more easily applicable to health advisories.

Table 3. List of parameters determined approximately monthly at the proposed monitoring station at Middle River (MDM). Funding from this proposal will cover cyanotoxin analysis for 18 sampling dates (18 dates, plus replicates and blanks), and analyses of other parameters not covered by other efforts.

Parameter	Approx. # Samples ($ this study)	*Approx. # Samples ($ other)*	Information Provided
Nitrate (NO3-N) (µM) **Nitrite (NO2-N)** (µM)	8	*14*	nitrogen as nitrate available for biological uptake; laboratory measurement to verify and calibrate in-situ data, increases due to nitrification or new inputs, decreases due to uptake and denitrification
Ammonium (µM)	8	*14*	nitrogen as ammonium available for biological uptake; tracer of wastewater source; shown to impact phytoplankton abundance, species composition, and primary production; increases due to mineralization or inputs decreases due to nitrification and uptake
Total Dissolved Nitrogen (TDN) (µM)	8	*14*	total nitrogen in the dissolved phase used to track the total N budget
Dissolved Organic Nitrogen (DON) (µM)	8	*14*	includes only the dissolved organic nitrogen fraction, used to track the total N budget; tracer of water source: Calculated as TDN-NO3-NO2-NH4
soluble reactive phosphate (SRP, PO4) (µM)	8	*14*	required nutrient for phytoplankton; has been shown to be inhibitory at high concentrations; tracer of water source
Chlorophyll-*a* & Phaeophytin (mg L^{-1})	0	*14*	laboratory measurements to verify and calibrate in-situ fCHLA data; phaeophytin to chlorophyll-a ratio provides information about algal growth versus senescence; tracer of water source
Phytoplankton Enumeration (cells L-1 and cm^3 L-1 by species)	8	*14*	microscope analysis for phytoplankton species identification, counts and biovolume; provides information about phytoplankton abundance and species composition; identifies whether the phytoplankton pool is made up of beneficial or harmful species; indicator of nutritional quality of the phytoplankton pool
Picocyanobaceria (cells L^{-1} and cm^3 L^{-1})	8	*14*	epifluorescence analysis that identifies picocyanobacteria (< 2 microns); identifies fraction of the phytoplankton pool that is made up of small cyanobacteria that are believed to be less favorable to the health of the food web
Cyanotoxins Whole Water (µg L^{-1}) SPATTs (ng g^{-1} day^{-1})	 22 22	--	**LCMS-MS** analysis for the detection of Anabaenopeptins, Anatoxin-a, BMAA, Cylindrospermopsin, Microcystins, Nodularins, and Saxitoxins
Cyanotoxins Whole Water (µg L^{-1}) SPATTs (ng g^{-1} day^{-1})	 8 8	--	**ELISA** analysis for the detection of microcystins, anatoxins, cylindrospermopsins, and saxitoxins

Task 2: Cyanotoxin monitoring at Stockton Waterfront

The Stockton Waterfront experiences dense and toxic cyanoHABs each year. Toxins in the samples have exceeded the highest recreational health advisory levels established by the State Water Board. While cyanotoxin monitoring has occurred at USGS and DWR water quality stations, cyanoHAB hot-spots in the Delta, such as Stockton Waterfront, have rarely been monitored. Because a research team, led by Dr. Hans Paerl and funded by the DSP, will be studying cyanoHAB aerosols at Stockton Waterfront in 2022, a unique opportunity exists to collect additional samples and leverage the presence of Dr. Paerl's team to understand bloom dynamics at the Stockton Waterfront.

Cyanotoxin sampling for public health focuses on the top 1 meter of the water column, but the full water column often must be considered when seeking to understand the factors driving a bloom. The changing vertical environment from surface to riverbed has rarely been studied in the Delta.

We propose collecting vertical profiles of the bloom by deploying a bbe Fluorometer and other continuous monitoring devices on a deployable cage (Figure 4; Table 4). We will also collect discrete water samples at multiple depths (Table 5). Pairing Fluoroprobe profiles and discrete samples will provide high-resolution data about the vertical distribution of cyanobacteria with discrete water data that can be compared with to management objectives for different water quality parameters. Data will be collected 6 times – approximately monthly May-October (start date depends on when bloom develops) – at three sites spanning a gradient of tidal and mixing energy (Figure 5). We hypothesize that the different mixing energy from tides and flow will change bloom structure and vertical mixing. Multiple profiles will be collected over ~6 hours on each sampling date to collect data on flood, slack, and ebb tides.

By sampling from surface to sediment, we will generate cyanotoxin data that can be used for public health assessments and nutrient data at depth that will be useful in characterizing the phytoplankton and nutrient environment of the Stockton Waterfront. The low dissolved oxygen levels in Stockton may result in nutrient release from the sediments, which could be supporting high cyanoHAB biomass. These data will provide information about the vertical and horizontal spatial heterogeneity in bloom density and other environmental parameters to better understand the factors driving and sustaining cyanoHABs at the Stockton Waterfront.

Figure 4. Vertical profling cage being prepared for deployment.

Table 4. Instruments installed on vertical profiling cage.

Instrument	Measurement description
Bbe Fluoroprobe	Abundance of different phytoplankton organisms: Cyanobacteria, Diatoms, Green Algae, and Cryptophytes
Sea Bird Wetstar	Chlorophyll-a
Sea Bird turbidmeter	Turbidity
PAR sensors	Photosynthetic Active Radiation (PAR): the light available for photosynthesis
Sea Bird AC-S	Absorption and scattering properties of water
Fastcat CTD	Conductivity, temperature, salinity, and depth
Underwater camera	Prototype sensor to estimate bloom biomass at low densities

Table 5. Discrete water quality parameters collected during vertical profiling. Samples will be collected on 6 dates (approx.. monthly from May-October) at 3 sites and 2 depths at each site (6 * 3 * 2 = 36 samples plus 8 QA/QC = 44 samples).

Parameter
Cyanotoxins whole water (µg L^{-1})
Nitrate (NO3-N) (µM) **Nitrite (NO2-N)** (µM)
Ammonium (NH4-N) (µM)
Total Dissolved Nitrogen (TDN) (µM)
Dissolved Organic Nitrogen (DON) (µM)
Soluble Reactive Phosphate (SRP, PO4) (µM)
Chlorophyll-*a* & Phaeophytin (mg L^{-1})
Phytoplankton Enumeration (cells L-1 and cm3 L-1 by species)
Picocyanobaceria (cells L^{-1} and cm^3 L^{-1})
Optical Properties (absorbance, fluorescence) (intensity)

Figure 5. Map of Stockton Waterfront showing potential profiling locations with green markers.

Task 3: Cyanotoxin collection during High-Frequency Mapping Surveys

The Delta is a mosaic of different channel and habitat types. To sample across spatially heterogeneous environments, the USGS has conducted Delta- wide high-frequency mapping surveys in Spring, Summer and Fall of 2018, 2020 and 2021. These boat-based mapping surveys cover approximately 350 miles of the Delta over 4 consecutive days. The underway flow-through sampling system allows us to simultaneously collect in situ data for water quality using a YSI EXO (water temperature, specific conductance, turbidity, pH, DO, total chlorophyll [fCHL], phycocyanin [fPC], and fluorescent dissolved organic matter [fDOM]), dissolved nutrients (nitrate using a SUNA nitrate sensor and ammonium using a

modified flow through Timberline Analyzer™). Phytoplankton abundance and taxonomic groups are assessed using a bbe Moldaenke™ Fluoroprobe that differentiates cyanobacteria, diatoms, green algae, and chlorophytes based on their characteristic pigments (Table 1). During these surveys discrete samples are also collected at ~30 fixed stations (Figure 6) for nutrient analyses (total and dissolved forms of nitrogen (nitrite, nitrate, ammonium, organic nitrogen) and phosphorus (phosphate), microscopic enumeration of phytoplankton, and direct quantification of the picocyanobacterial abundances using epifluorescence microscopy (EFM).

These Delta wide mapping surveys were funded by the Delta RMP in 2018 and the Delta Science Program (DSP) in 2020 and 2021. In summer 2020 we secured internal USGS matching funds to add-on cyanotoxin monitoring (whole water and SPATT) to these surveys. In 2022 the three surveys will be funded by the State Water Contractors (~$450,000), however funding to collect cyanotoxins during these mapping surveys is not identified in 2022. We propose that the Delta RMP fund the collection of cyanotoxins during 1, 2 or 3 mapping surveys in 2022. This will allow us to relate cyanotoxin data to the rich suite of parameters already being collected.

Task 3. Option A: We will collect discrete whole water samples for analysis using a combination of the methods of LCMS-MS (Liquid Chromatography-Mass Spectrometry) and ELISA (Enzyme-Linked Immunosorbent Assay). Samples will be collected from ~0.3 meter depth at 30 fixed stations across the Delta. Whole water samples will be placed immediately on dry ice in the field, kept frozen (-80° C), and then sent to the laboratory (Lumigen Instrument Center) for analysis. All (100%) whole water samples will undergo analysis via LCMS-MS and – upon review of LCMS-MS data – a subset of samples (~20%) will be selected for analysis via ELISA (BSA Environmental Services).

Task 3. Option B: In addition to collecting discrete samples during mapping surveys, we can also integrate SPATT samplers into our flow-through system on the boat to generate a continuous estimate of cyanotoxin concentrations across the Delta (Figure 7). Cyanotoxins will be measured while underway by passing water from our onboard system over the SPATT samplers, which will provide a spatially integrated measure of dissolved toxins. SPATTs will kept in place as the boat covers ~5-10 km reaches between stations. The spatial extent each SPATT sample is associated with will be informed in real time by on-board measurements, visual observations of the water, and information gleaned from prior studies. Approximately 30 SPATTs will be collected during each mapping survey including quality-control blanks and replicates. Because the of the additional cost of SPATT samplers, this component is being separated out as a separate and optional sub-task

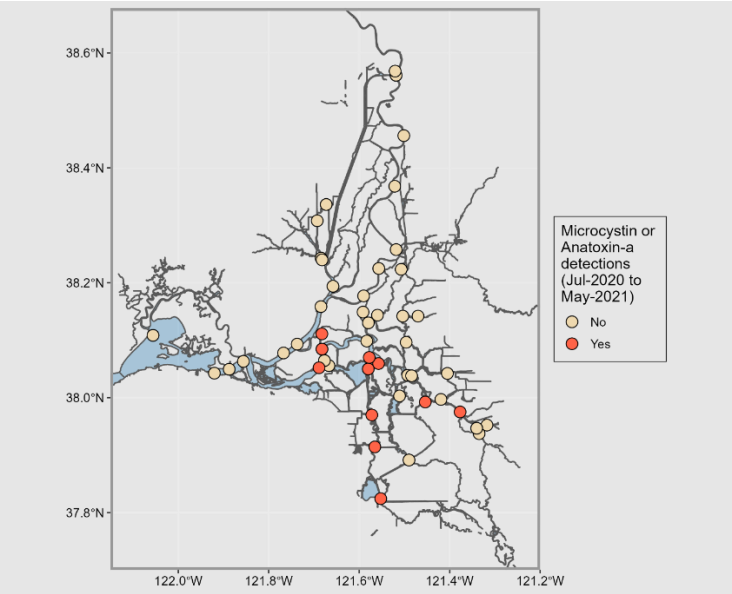

Figure 6. Detection of microcystin or anatoxin-a at fixed stations during high-frequency mapping campaigns conducted in July 2020, August 2020, October 2020, and May 2021. Samples from summer and fall 2021 have not been analyzed yet.

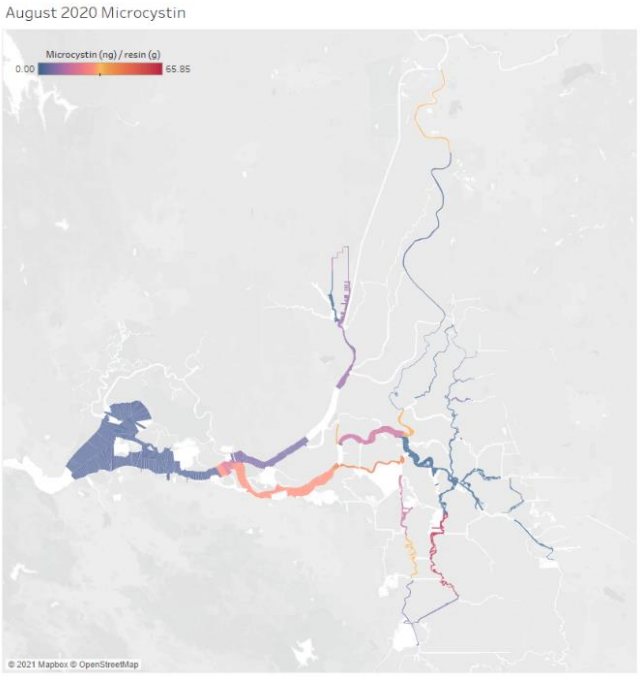

Figure 7. Map of SPATT microcystin levels from the flow-through SPATT deployed during the mapping survey in August 2020. The SPATT provide a spatially integrated method to detect cyanotoxins in the Delta.

Project Timeline and Deliverables

- **Project Start-End Dates: March 1, 2022 through December 31, 2023.**
- Samples collection (whole water and SPATT) will occur over a 12-month period, starting in early 2022.

- Cyanotoxin data will be made available within 6 months following data collection and analysis via the USGS and CDWR database systems, or upon request. These data will also be made available using online visualization tools

  (e.g., https://tableau.usgs.gov/views/Bay_Delta_Portal/Portal?:embed=yes)
- Results will be reported to the Delta RMP, local conferences (e.g. Bay Delta, IEP), and upon request.
- A report that describes the approach and methods, summarizes any issues or lessons learned that occurred during data collection, provides tabular and/or graphical summaries of the spatial and temporal patterns in the data, evaluates the data quality, and relates study findings to the Delta RMP management questions will be provided at the end of the agreement. The report will also include comparison between the whole water and SPATT data and between the LCMS-MS and ELISA data.
- We anticipate data from this study along with other relevant data collected by the USGS and DWR will be incorporated into a journal article, IEP Newsletter article, and/or USGS report.

Budget

The total amount requested from the Delta RMP is **$77,189** for Task 1, **$108,864** for Task 2, and **$36,954 or $61,639** for Task 3 (Table 6). Task 3 is budgeted as the cost of a single mapping survey, and Delta RMP could choose how many of the 3 planned surveys in 2022 they would like to add cyanotoxin sampling to.

This will cover USGS staff time and associated costs (e.g., boats, vehicles, fuel, supplies, instrument costs, travel, chlorophyll and nutrient analyses, phytoplankton enumeration). This budget assumes the Delta RMP will contract directly with BSA and Lumigen to cover analytical costs for cyanotoxins.

In Kind Contributions: Well over $750,000 (over $150,000 per station) in annual cost sharing will be provided by the USGS and DWR to support monthly field visits (staff time, boats, vehicles, fuel, sampling equipment), analytical costs associated with samples listed in Table 2 that are collected monthly at these stations, and collection of in situ continuous monitoring data at the four stations.

Table 6. Budget for each of the three tasks described in this proposal.

Proposed Tasks	Costs
1. Cyanotoxins at MDM	
Cyanotoxin analyses*	$23,580
Water quality analyses (e.g., nutrients, phytoplankton enumeration)	$5,633
USGS personnel, boats, vehicles, supplies, equipment, etc.	$58,275
Total project cost	$87,488
USGS match	$10,299
Cooperator cost	**$77,189**
2. Stockton Waterfront	
Cyanotoxin analyses*	$8,640
Water quality analyses (e.g., nutrients, phytoplankton enumeration)	$34,214
USGS personnel, boats, vehicles, supplies, equipment, etc.	$77,905
Total project cost	$120,759
USGS match	$11,895
Cooperator cost	**$108,864**
3.A Cyanotoxins during Mapping Surveys no SPATT (1x)	
Cyanotoxin analyses*	$17,280
Water quality analyses (e.g., nutrients, phytoplankton enumeration)	$0
USGS personnel, boats, vehicles, supplies, equipment, etc.	$24,592
Total project cost	$41,872
USGS match	$4,918
Cooperator cost	**$36,954**
3.B Cyanotoxins during Mapping Surveys with SPATT (1x)	
Cyanotoxin analyses*	$38,520
Water quality analyses (e.g., nutrients, phytoplankton enumeration)	$0
USGS personnel, boats, vehicles, supplies, equipment, etc.	$28,899
Total project cost	$67,419
USGS match	$5,780
Cooperator cost	**$61,639**

*Costs associated with cyanotoxin analyses will be paid directly to BSA and Lumigen.
**Budgets were developed assuming the USGS agreement is routed through an entity that can receive USGS cooperative matching funds.

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