

**Marine Pollution Bulletin**  
**Initial estuarine response to the nutrient-rich Piney Point release into Tampa Bay,**  
**Florida**  
--Manuscript Draft--

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<b>Abstract:</b>	Legacy mining facilities pose significant risks to aquatic resources. From March 30th to April 9th, 2021, 814 million liters of phosphate mining wastewater and marine dredge water from the Piney Point facility were released into lower Tampa Bay (Florida, USA). This resulted in an estimated addition of 186 metric tons of total nitrogen, exceeding typical annual external nitrogen load estimates to lower Tampa Bay in a matter of days. An initial phytoplankton bloom (non-harmful diatoms) was first observed in April. Filamentous cyanobacteria blooms ( <i>Dapis</i> spp.) peaked in June, followed by a bloom of the red tide organism <i>Karenia brevis</i> . Reported fish kills tracked <i>K. brevis</i> concentrations, prompting cleanup of over 1600 metric tons of dead fish. Seagrasses had minimal changes over the study period. By comparing these results to baseline environmental monitoring data, we demonstrate adverse water quality changes in response to abnormally high and rapidly delivered nitrogen loads.
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Dr. Michael Boufadel, Dr. Francois Galgani, Dr. Gui-Peng Yang  
Co-Editors-in-Chief  
Marine Pollution Bulletin

We are pleased to resubmit our manuscript, "Initial estuarine response to the nutrient-rich Piney Point release into Tampa Bay, Florida" to be considered as an original research article in Marine Pollution Bulletin.

An earlier draft of this paper was submitted in December of last year and we received comments from three reviewers. Although the initial decision was to reject the manuscript, we contacted Dr. Boufadel about the quality of one of the reviews and we were encouraged to revise the draft for resubmission. We have carefully considered comments from all three reviewers of the original submission and have made substantial revisions in response to these comments. Overall, we have made many edits to improve clarity of the text and have shortened the content in many locations. The point-by-point responses are detailed below and we have included a version of the revised manuscript with track changes to aid in the review. We are confident these changes have addressed the concerns of our reviewers.

The impact of this event on Tampa Bay is important both regionally and nationally to raise awareness of how insufficient regulatory oversight and planning can lead to unintended environmental impacts. We stand by our conviction that these events need to be documented in the primary literature to advance the conversation on how legacy mining facilities can be safely and responsibly closed. We appreciate the opportunity to publish this important work in Marine Pollution Bulletin.

Sincerely,

A handwritten signature in black ink, appearing to read "Marcus W. Beck".

Dr. Marcus W. Beck  
Program Scientist  
Tampa Bay Estuary Program

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TAMPA BAY ESTUARY PROGRAM

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U.S. ENVIRONMENTAL PROTECTION AGENCY.

We sincerely thank the reviewers for providing comments on our manuscript. We provide a point-by-point response to these comments below. In general, most of the revisions address concerns raised by all three reviewers, including 1) numerous edits to improve the writing for clarity and brevity, and 2) additional information on sampling methods. We have also addressed concerns of Reviewer 2 about potential nutrient cycling by adding and expanding a section in the discussion and concerns of Reviewer 4 regarding additional detail on the simulation modelling.

All page and line numbers in the reviewer comments refer to the original submission. Page numbers noted in our response refer to the revised submission.

## Reviewer 1

The authors (a very long list of authors) have described the sequence of events that followed a phosphate mining spill. The paper is long and tedious- and in the end cannot conclude any strong relationships with red tide occurrence. I wish I could be more positive about this paper, but it has the feeling that authors were rushing to get something in press, rather than taking the time to write a well crafted manuscript. There are some very good writers on the list of authors- so I am a bit surprised that they all agreed to this final version.

- **Response:** Thank you for your comments. We have made revisions to improve the writing, by reducing length in many locations and making additional edits where noted. Many of the edits in response to the specific comments from the other reviewers are used to address some of the comments below.

graphic abstract- much too complicated; fonts too tiny, too much text

- **Response:** The graphical abstract was simplified and the font size increased.

highlights- the term Piney Point is meaningless except for locals

- **Response:** The first highlight was changed to “186 metric tons of total nitrogen from wastewater were added to Tampa Bay.”

abstract- poorly written

- **Response:** Abstract was edited.

Introduction- needs a complete rewrite- it takes more than 3 pages to get to the point that there was a spill. The general background is interesting but you need to grab the readers attention first.

- **Response:** Introduction was shortened and edits were made to improve clarity.

p. 5- inadequate explanation for why the focus on N. Why not both?

- **Response:** Please see our response to a similar comment from reviewer 4. Many studies, as well as the successful nutrient management paradigm for Tampa Bay, have demonstrated that Tampa Bay is nitrogen-limited. Phosphorus trends are also provided in the supplement.

Methods- the first paragraph on monitoring is all about modeling, not monitoring

- **Response:** We have added a “Modeling” sub-section at the beginning of the methods.

p. 7- how were the Karenia data quantified...they didn't just appear in a database.

- **Response:** Text was added: "HAB sampling typically occurs in response to bloom events or fish kills with extensive quality control of cell counts conducted by FWC-FWRI (additional details in [Stumpf et al. 2022](#)). HAB data were restricted to Tampa Bay boundaries and over 90% of the samples were collected within one meter of the surface."

p. 7 - eyes on seagrass is meaningless (except for locals)

- **Response:** We have removed this from the text.

stats section- poorly written, confusing, wordy

- **Response:** Please see the responses to reviewer 2.

Results- rambling, disorganized and non-quantitative. Paragraphs don't make sense (try using topic sentences and structuring concepts accordingly)

- **Response:** Please see the responses to reviewer 2, particularly regarding the first section of the results.

Discussion- I was left wondering what I learned. No conclusions drawn

- **Response:** Please see the responses to reviewer 2, particularly regarding the edits to the discussion sections.

In all, it seemed like an interesting exercise to describe all the patterns, but this does not make a quality manuscript that will stand the test of time.

- **Response:** We are confident that the edits made in response to your comments and those from the other reviewers have made this manuscript a more compelling and valuable contribution.

## Reviewer 2

Beck and colleagues report on the short- and mid-term effect of a very large nutrient-rich release from Piney Point. The authors focus on a few water quality indicators, but also include data for several additional responses (e.g., seagrass coverage). The results are largely what you would expect, initial rapid response of phytoplankton with cascading secondary responses.

- **Response:** We really appreciate your comments. Please see the detailed responses below.

First, a few of the things that make this a worthwhile contribution to MPB. The authors should be commended for their quick response to data collection and the large number of data streams that were brought to the study. It really is quite impressive. Further, compared to citizen monitoring, I expect the data is high quality and consistent. Second, the writing was quite good; there were some sections where flow or organization could be improved (beginning of results; last section of discussion); however, these considerations are very minor.

- **Response:** Thank you for pointing out the merits of this paper and providing suggestions where the writing can be improved. We have edited the sections you point out for brevity and clarity.

Second, a few things that could be improved or warrant some critique. What is the rationale for the lack of continuous line numbers? Journal suggestion? Reviewing is so much easier when line numbers line up with the text and continue throughout the document. I realize this is not something that should be fixed now, but it is bothersome.

- **Response:** Unfortunately, the line numbers are generated automatically by the journal submission system and we have no control over how they are presented. We appreciate your use of both page and line numbers in your comments to reference text locations.

That said, my one major critique of the manuscript, one that is difficult to reconcile with the available data, is the speculative nature of last section of the Results, titled Potential nutrient cycling. This is clearly a very important section and, in my opinion, warrants additional consideration. The title of this section is correct, yes; thus, I would like to see 1) this section moved to the discussion, 2) the MAJOR lines of evidence supporting this thesis outlined and discussed consecutively in one section, 3) major alternatives entertained and addressed. The authors might also think about what it would take to make the connections more explicit, if possible (stable isotopes, modeling, etc.).

- **Response:** Thank you for this comment. We agree this section is important, although it is definitely speculative without more quantitative results that definitively link the pieces. As such, we have moved the content to the discussion and expanded to text to include a more nuanced description of the trends, including what information may be needed to support the narrative. This includes information from the final paragraph of the discussion, which was shortened and combined with the prior paragraph. The new section begins on page 18.

#### MINOR COMMENTS/SPECIFIC SUGGESTIONS

P3/L56: Recommend removing “in other countries.” When I read that passage it seems to suggest a broader critique, one that is more political. That may not have been your intention, but that was how I read it.

- **Response:** Our point was to suggest that mining products in Florida are generally exported and have little benefit for the local communities. However, this may be a bit editorialized and was removed.

P4/L21-24: I am not sure if “unanticipated releases” captures the spirit in which these releases occurred. In fact, the title of the FWC report was “Response of estuarine nekton to the regulated discharge of treated phosphate-production process water.” This regulated discharge seemed to occur after extensive consultation with the EPA.

- **Response:** Agreed, we have removed “unanticipated.” We agree that the releases often occur after consultation or approval from regulatory bodies, but they are generally unanticipated in that long-term site management has not planned for these events, nor the potential impacts on the environment.

P4/L51: worth indicating that these concentrations, especially for TP, are several orders of magnitude greater than typical surface waters?

- **Response:** The sentence was edited: “Water quality of NGS-S measured in 2019 showed total phosphorus (160 mg/L) and total nitrogen (230 mg/L) were approximately three orders of magnitude higher than typical concentrations in Tampa Bay.”

P4/L56: deliberately “released”

- **Response:** Changed to “From March 30th to April 9th, approximately 814 million liters (215 million gallons) of stack water were released to lower Tampa Bay following an [emergency order](#) authorized by the Florida Department of Environmental Protection (FDEP).” A similar, redundant sentence earlier in the paragraph was removed.

P5/L4: I see the use of the term lower Tampa Bay; how does that compare to Figure 1, the areas of interest? Ultimately, lower Tampa Bay is used extensively, but I don’t see it defined. Also, is this region ecologically based or defined by currents?

- **Response:** The boundary between middle and lower Tampa Bay was added to Figure 1. These boundaries are used by the Tampa Bay Estuary Program to track annual attainment of water quality targets under a Reasonable Assurance plan for TMDL reporting to the Florida Department of Environmental Protection. We do not define the rationale for the boundaries in the main text because the regulatory delineations of the bay are not of interest to a broader audience, but we use the delineation as it is useful for our discussion of trends in the paper. Our area delineations (areas 1 - 3) were meant to provide a more relevant grouping for the paper.

P5/L22-27: I think a citation may be warranted here; while predictable, numerous studies ultimately supported the statement that nitrogen is limiting in Tampa Bay.

- **Response:** Agreed, we have revised this statement to include additional citations and explanation:

“Numerous studies, as well as the successful nutrient management paradigm, have demonstrated nitrogen-limitation in Tampa Bay and the system is generally considered phosphorus enriched ([Wang, Martin, and Morrison 1999](#); [Greening and Janicki 2006](#); [Greening et al. 2014](#)). As such, we focus on nitrogen in our analyses as the identified limiting nutrient for Tampa Bay and its potential to create water quality conditions unfavorable for seagrass growth due to enhanced algal production.”

P5/L29: interesting that discharge was to Port Manatee and not Bishop Harbor like the release discussed previously.

- **Response:** Yes, we believe the intent was to minimize impacts to sensitive back-bay habitats that were observed in previous releases, although we have no information to verify this claim and have not included it in the text.

P6/L24-39: that is an impressive and extensive list of agencies and partners; any potential drawbacks to so many data streams?

- **Response:** Data quality assurance and control was a primary concern with multiple partners collecting information. We have included additional information about the data collection procedures and how the Tampa Bay Estuary Program worked to facilitate quality of the data. Please see the additions starting on page 6.

P6/L54-65: is it routine to take water samples directly from the surface? Ultimately, were the authors worried about any potential stratification, particularly of Karenia brevis.

- **Response:** Most of the water quality and phytoplankton samples were surface collections following standard sampling methods used by our partners (see the text addition about sampling). Tampa Bay also has a very shallow average depth of just over 3m and is not strongly stratified for most of the year. So, we feel confident that the majority of samples used in our analyses were generally representative of conditions in the water column. Anecdotally, benthic fishes were observed during the fish kills (e.g., eels, rays), so it is likely that *K. brevis* was present throughout the water column, although we have very little quantitative data to describe the vertical distribution. Please see the addition of information on *K. brevis* sampling at the top of page 7 in response to reviewer 1.

P7/L32-37: why get data from Tampa Airport and the Airfield in St. Petersburg? There is an NWS location not far from Piney Point.

- **Response:** We have updated our weather data to include wind and precipitation from Albert Whitted Field in St. Petersburg and have excluded the precipitation data from Tampa International Airport. Our initial rationale for using data from Tampa was a longer period of record. However, we have revised the historical baseline in the plot to include only years 2006 to 2020, consistent with our comparison of the water quality data to historical observations. Also, the NWS location is in Ruskin, Florida and is farther from the bay proper than the location in St. Petersburg.

P9/L27-29: not sure what to make of the aggregating data to weeks or months; isn't there a way to maintain the actual sampling regime?

- **Response:** The weekly aggregation was only done to allow plotting and comparison between variables on a common scale (e.g., Figure 6 and 8 in the revised text). None of the statistical tests used data with fixed sample dates. We removed the text because the statement is inaccurate and somewhat misleading.

P9/L49: This is a bit confusing; is this comparing “between” pre and post?

- **Response:** No, these tests were meant to compare all observations post-release to help identify periods of time when observations differed, e.g., when chlorophyll was at its maximum. The text was revised for clarity.

“Differences in observations between months for April to September for water quality, seagrass, and macroalgae within each area (Figure 1a) were evaluated using a Kruskal-Wallis one-way analysis of variance (ANOVA) followed by multiple comparisons using 2-sided Mann-Whitney U tests ([Hollander, Wolfe, and Chicken 2013](#)). These tests were used to statistically characterize the temporal progression of changes in the bay following release from Piney Point, e.g., were July conditions significantly different from April?”

P11/L12-14: as personal preference, I see no reason to lead off the results with a general statement about the location of a figure summary. Instead, the authors might reference the figure when they describe, in general, that a bloom was observed ~2 weeks after the release began, red tide was observed within x weeks, the bloom peaks, and conditions become stable on xx.

- **Response:** The first section of the results describing the timeline was removed and text was placed in the appropriate sections that followed. The timeline is now referenced in the first paragraph of the discussion to provide a synthesis of the main results that were described.

P11/L20-: this first part of the results has a choppy structure

- **Response:** Please see the response to the previous comment.

P11/L31-34: the authors indicate the Tampa Bay influx was related to an ongoing coastal bloom; is this indicating the red tide bloom was ongoing prior to the Piney Point discharge? Also, not clear what “related” means in this context. This important statement may require additional context.

- **Response:** Yes, red tide was observed in the Gulf of Mexico prior to the occurrence in Tampa Bay. Content in the red tide section was edited to make this clear (page 15):

“This first Tampa Bay influx originated from an ongoing coastal bloom in the Gulf of Mexico, as is common when red tide is observed in the bay ([Steidinger and Ingle 1972](#); [Flaherty and Landsberg 2011](#)).”

Blooms in Tampa Bay originate from *K. brevis* that is transported in from the Gulf of Mexico. An important distinction that we make in this paper is that, although blooms in Tampa Bay are not uncommon, the severity of the 2021 bloom was very likely related to the favorable conditions in the bay prior to July. These favorable conditions were described in the paper as 1) nutrient availability from Piney Point, and 2) high salinity conditions from low rainfall. The passage of tropical storm Elsa was also a confounding factor. It is nearly impossible to identify a single factor causing the observed conditions, but we describe the considerations in the discussion (page 23).

P16/L26: not sure I see the rationale for analyzing the data using Braun Blanquet estimates, which, in my impression, was developed as a rapid assessment

- **Response:** Seagrass transect monitoring in Tampa Bay has occurred annually since the mid-1990s using similar methods as in this paper (i.e., [E. Sherwood et al. 2017](#)). The primary difference for the annual monitoring is the transect length, which is typically much longer than the 50m used in the 2021 rapid design. This shorter distance was chosen to allow for quicker sampling times and greater coverage in response to the rapidly changing conditions in 2021. Thus, the rapid design provides data that are similar quality to the long-term data, with notable exceptions indicated in the manuscript (i.e., no seasonal estimates). Our rationale for evaluating Braun Blanquet coverage was to verify the results obtained evaluating frequency occurrence estimates. The text was modified to make this clear (page 15):

“Tests using Braun Blanquet cover estimates confirmed the results from the frequency occurrence estimates (Tables S3, S4).”

P17/Potential nutrient cycling: Unfortunately, much of the latter portions of this section are speculative and better presented in the discussion. Further, it would have been nice to see something a bit more concrete linking the nutrient shift from photoplankton to macroalgae and release of macroalgae nutrients to *K. brevis*; stable isotopes perhaps; the nitrogen sources could have been highly enriched

- **Response:** This section was moved to the discussion, please see the response to your general comment above.

P18/L18: or a shift in the relative availability, N:P

- **Response:** Added text “...or changing availability of nutrient ratios creating favorable conditions for macroalgae growth ([Valiela et al. 1997](#); [Cohen and Fong 2006](#)).”

## Reviewer 4

This is an interesting study in that it utilizes a quasi-Lagrangian approach to quantify the influence of a nutrient-rich waste plume on an estuarine ecosystem and is the first that I am aware of that documents the impacts of the Piney Point episode. Nonetheless, there are some points that require clarification to give the reader confidence in the findings. In general, more details are needed on several aspects of the methodology, and I provide specific details below on this as well as other issues.

- **Response:** Thank you for your constructive comments. Please see the detailed responses below.

Page 1, Line 38 - “Elevated levels of phytoplankton” - needs to be clearer... is this referring to biomass or abundance?

- **Response:** This line was changed to “An initial phytoplankton bloom (non-harmful diatoms) was first observed....” This was based on chlorophyll measurements in the vicinity of Port Manatee and cell concentrations enumerated to taxa from laboratory analysis. The word limit for the abstract prevents these details, but they are presented in text (page 18).

Pages 6, Lines 4-22 - The sampling regime was largely dictated by output of a model of plume evolution. It would be useful to see more details on how the model is calibrated, and if/how its output is cross-verified. In other words, are field observations of T/S compared with model output to evaluate its performance in terms of accuracy of plume location? This seems like critical information for assuring that the sampling regime actually followed the plume.

- **Response:** TBCOM was previously tested for veracity against in situ observations (e.g., [Chen et al. 2018](#)). A prior version upon which TBCOM was constructed was also veracity tested and used in several studies (Zhu et al., 2015a,b,c, and [Chen et al. 2019](#)) More systematic model/data comparisons were also reported in a recent PhD dissertation (J. Chen, Univ. of South Florida, 2021). Thus, prior to this application, TBCOM was found to perform well in simulating the circulation in the Tampa Bay, and TBCOM has provided publicly available, daily nowcast/forecasts (<http://ocgweb.marine.usf.edu/~tbo/index.html>) for the past four years.

More detailed model description or analysis is beyond the scope this work. A dedicated manuscript is being prepared to document the TBCOM tracer model as a rapid response to the Piney Point event. Preliminary results can be found in CERF and AGU presentations:

Liu, Y., Weisberg, R.H., Zheng, L., Sun, Y., Chen, J. (2021), Nowcast/Forecast of the Tampa Bay, Piney Point Effluent Plume: A Rapid Response, Abstract (OS35B-1036) presented at AGU Fall Meeting, New Orleans, Louisiana, December 2021.

Liu, Y., Weisberg, R.H., Zheng, L., Sun, Y., Chen, J. (2021), Nowcast/Forecast of the Tampa Bay, Piney Point Effluent Plume: A Rapid Response, Coastal and Estuarine Research Federation (CERF) 2021 Virtual Meeting, November 2021.

The main text was edited to include some of the above details (page 5): “The modeled plume evolution web product (<http://ocgweb.marine.usf.edu/~liu/Tracer/>) served as the principal guidance for coordinating the data collection during the event. Preliminary model results for Piney Point are reported in [Liu et al. \(2021\)](#) and previous model veracity testing was described in [Chen et al. \(2018\)](#) and [Chen et al. \(2019\)](#) (and references therein)”

Page 6, Lines 46-54 - More information is needed on laboratory procedures. In particular, with many different entities running samples and presumably utilizing different analytical approaches for each variable, it is important to know how comparable the data is between programs.

- **Response:** Our initial submission included supplement material that described the sample collection and processing in more detail. We have moved that content from the supplement to the main text, beginning on page 6:

"Most samples were surface collections by boat, with sample frequency approximately biweekly for locations around Piney Point, although effort varied by monitoring group and was more consistent during the first three months after the release. Established laboratory and field sample protocols for all survey methods were based on an [Interagency Monitoring Project Plan](#) maintained by the TBEP and those of the inter-agency partners. Data quality objectives followed guidelines outlined in the USEPA-approved TBEP Data Quality Management Plan ([E. T. Sherwood et al. 2020](#)). Many of the local partners also participate in the Southwest Florida [Regional Ambient Monitoring Program](#) (RAMP) that ensures similar standards and protocols are followed in the collection and processing of monitoring data, including routine cross-reference of split samples between laboratories to check precision of measured values. Samples requiring laboratory analysis (e.g., nutrient assays) were obtained primarily from bottle collection at the surface, whereas in situ measurements were available for many parameters (e.g., dissolved oxygen, Secchi depth, etc.). In situ measurements were collected using common monitoring equipment, such as YSI sondes or Seabird CTD casts, depending on monitoring agency. Laboratory methods used to process samples were based on accepted procedures promoted through the Southwest Florida RAMP."

Page 7, Line 22 - Were the breakpoints for K. brevis, or total phytoplankton abundance?

- **Response:** These were applied to K. brevis. The sentence was revised: "Bloom sizes for HAB species were described qualitatively as low/medium/high concentrations..."

Page 7, Line 37 - What about from ungauged flows?

- **Response:** Our methods for estimating inflows into Tampa Bay are based on those similarly presented in a technical report published by the Tampa Bay Estuary Program ([Janicki Environmental, Inc. 2012](#)). This report provides the basis for compliance assessment of our TMDL for reporting to the Florida Department of Environmental Protection and represents our best estimate at hydrologic load inputs to the bay. We have provided a citation to this report in the paper.

Page 11, Line 30 - Need to put Anna Maria Sound on the map. Likewise for Port Manatee.

- **Response:** Locations were added to the map.

Page 12-14, Water quality trends section - Very little attention is given to the role of the hurricane that hit the area in early July. Would be interesting to hear more about its effects on water quality in the study area.

- **Response:** As noted in the red tide section and discussion, Tropical Storm Elsa was a confounding factor when interpreting drivers of the red tide event. It is challenging to disentangle an isolated effect of the storm on the red tide and the same could be said for the broader suite of parameters that were used to assess water quality. For example, a clear chlorophyll spike in early July coincided with the red tide peak, which also coincided with passage of the storm. A lack of continuous monitoring in the bay also prevented a more comprehensive assessment of potential storm effects. However, water quality

conditions were changing rapidly from the red tide, preceding the arrival of the storm by several days, and we consider the water quality conditions at the time of passage to be driven primarily by the *K. brevis* bloom in the bay. We have added some additional text describing these interpretations in response to your comment below about dissolved oxygen and fish kills.

Page 14, Macroalgae and seagrass trends section - This appears to be one of the weaker components of the project, although it is outside of my expertise. My biggest concern is that it is not clear to me how useful "% occurrence" is. As far as I can tell, this is not equivalent to biomass or areal coverage. Perhaps I am mistaken? Regardless, it would be useful to see a better justification for this metric. Also, how did the 2021 data compare to historic spatial-temporal trends?

- **Response:** Frequency occurrence is a routine reporting metric used by the Tampa Bay Estuary Program to summarize the long-term annual transect monitoring (see [Johansson 2016](#); [E. Sherwood et al. 2017](#)). Additional text on page 10 was added to make this clear:

"Frequency occurrence estimates were used to evaluate macroalgae and seagrasses as a standard metric used in previous analyses in Tampa Bay ([Johansson 2016](#); [E. Sherwood et al. 2017](#))."

We realize that additional metrics could have been used to summarize the data and we presented trend assessments using the Braun-Blanquet abundance estimates as confirmation for the results from the frequency occurrence analyses (Tables S3 and S4). However, this does not provide an assessment of biomass or coverage. Weight data were collected for select macroalgae samples during the 2021 surveys and a preliminary analysis (see the Seagrass -> Biomass estimates tab [here](#)) suggested similar trends as the frequency occurrence results. Because we did not have comparable weight data for seagrasses, the analysis was not included. It is also worth mentioning that coverage estimates are obtained every two years for Tampa Bay as an annual snapshot (noted at the top of page 21 in the discussion), but these are not of sufficient temporal resolution to support the analyses in this paper.

There were limitations to our analysis that were noted in the original draft, primarily the lack of long-term historical data on seasonal trends for seagrasses and macroalgae. For example, page 22, line 52 in the original discussion: "Long-term monitoring data describing normal seasonal variation in macroalgae are unavailable and we cannot distinguish between seasonal and interannual changes and those in potential response to the Piney Point release."

Page 17, Lines 17-20 - How did the 2021 fish kills compare to historical kill spatial-temporal trends?

- **Response:** We were also interested in this question and had included a long-term trend plot in an earlier draft, with results as early as 1995 (see [here](#)). However, this was not included due to differences in reporting methods that have changed over time. Online reporting became available in recent years and we considered the results incomparable to the earlier data that were based on phone reporting methods.

Page 17, Lines 37-40 - Could there have been a role for low D.O. in the post-storm fish kills?

- **Response:** We included some text describing this issue in the discussion.

"Water quality conditions before and after passage of tropical storm Elsa may have also contributed to fish kills by reducing bottom-water dissolved oxygen. [Stevens, Blewett, and Casey \(2006\)](#) documented impacts of a category 4 storm on fish resources in the Charlotte Harbor estuary, although tropical storm Elsa was much

smaller and fish kills were documented prior to and after arrival of the storm. Lack of continuous monitoring data for bottom waters in Tampa Bay prevents a more detailed assessment of impacts of the storm on water quality."

Discussion - In general, it would be useful if the team would spend some time establishing the prevalence of N-limitation in the system, either based on their own data or previous studies. The interpretation of plume impacts is predicated on the system being N-limited, so that needs to be established up front.

- **Response:** We agree that the nutrient limitation needs to be clearly stated as the basis for focusing primarily on nitrogen and not phosphorus. The second paragraph in the introduction was meant to provide this justification, but this was clearly insufficient as noted in the review. We include some additional text and citations in the final paragraph of the introduction as further justification - see the response to the similar critique from reviewer 2. Also note that phosphorus summary plots were previously included in the supplement.

Page 20, Line 22 - Ammonium, not ammonia. Or was pH such that ammonia would be the dominant form?

- **Response:** This is in reference to total ammonia nitrogen, as was provided by the water quality sampling. We have made a note in the methods section (top of page 6) that any mention of ammonia refers to total ammonia nitrogen.

Page 20, Line 39 - Might be worth having a separate paragraph that talks about the timing/duration of the *K. brevis* bloom in 2021 compared to historic conditions.

- **Response:** Previous content in the discussion below this paragraph provided some historical context on red tide events in the region. Specifically, on page 23: "Occurrence of this species has historically been spatially distinct, with blooms originating in subsurface water offshore on the West Florida Shelf..."

We have added some text below the existing paragraph to provide more context: "Seasonal persistence in Gulf waters in southwest Florida can vary between years, with some blooms lasting as short as a few weeks, while others have been present for longer than a year (the 2018 bloom lasted sixteen months, [Skripnikov et al. 2021](#)). Severity in estuaries is typically less than Gulf waters as *K. brevis* is limited to higher salinity."

Page 20 (last paragraph)-21 (first paragraph) - Without showing methods and/or data, I feel that it is inappropriate to introduce the nekton abundance/composition here.

- **Response:** Agreed, much of this content was removed.

Page 21, Lines 32-47 - Much of this text on seagrass seems speculative, and again, unclear about use of % occurrence as a metric.

- **Response:** We have replaced the beginning of this paragraph with more definitive text:

"Previous research for Tampa Bay has identified water quality conditions that are likely to promote seagrass growth ([Greening and Janicki 2006](#); [Greening et al. 2014](#), and references therein). The results observed in 2021 suggested water quality conditions were not supportive of seagrass growth, although changes were not observed and the conditions likely did not persist long enough to impact seagrasses."

Also, please see our response above about frequency occurrence.

Page 24, Lines 17-52 - Speculative without showing methods of data collection or the actual data.

- **Response:** The paragraph was removed for brevity.

## References

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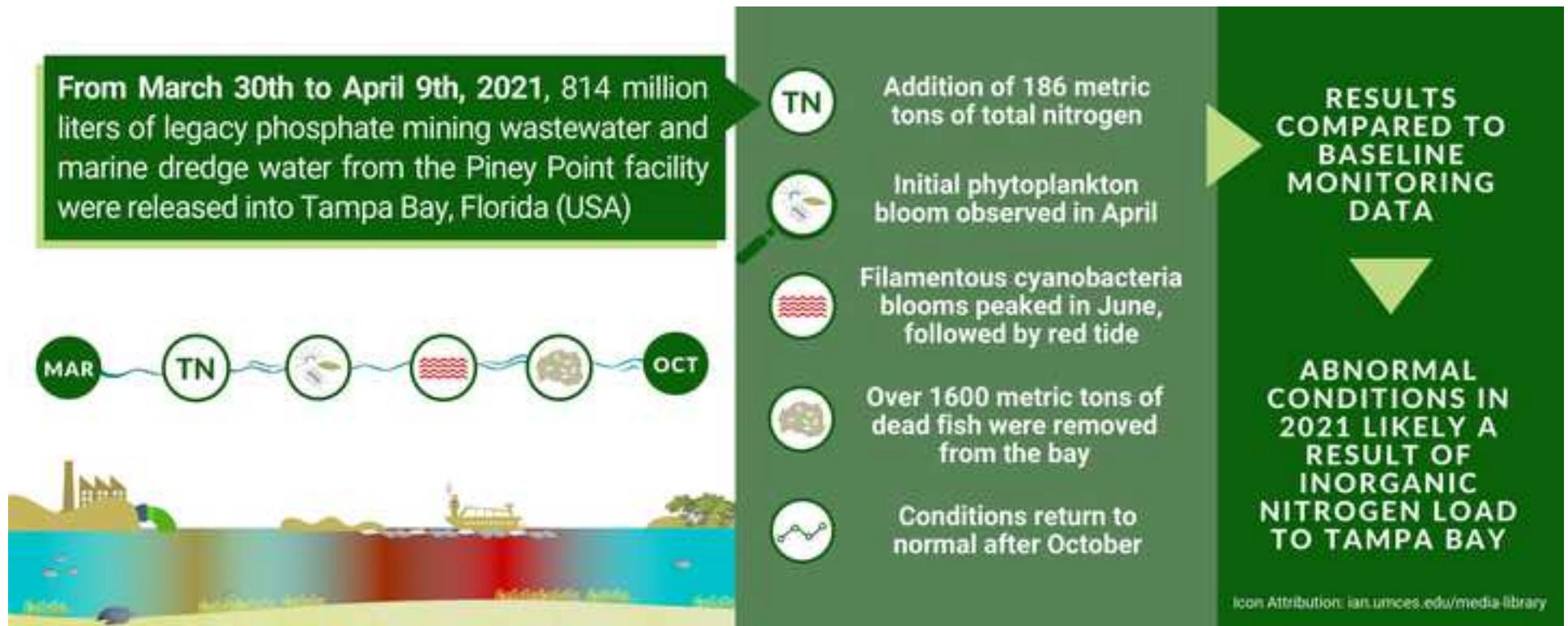
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- 186 metric tons of total nitrogen from wastewater were added to Tampa Bay
- An initial diatom bloom was observed near the release site
- Filamentous cyanobacteria were observed at high biomass
- *Karenia brevis* (red tide) was at high concentrations, co-occurring with fish kills
- Seagrasses were unimpacted during the six-month study period

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# Initial estuarine response to the nutrient-rich Piney Point release into Tampa Bay, Florida

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## Abstract

Legacy mining facilities pose significant risks to aquatic resources. From March 30th to April 9th, 2021, 814 million liters of phosphate mining wastewater and marine dredge water from the Piney Point facility were released into lower Tampa Bay (Florida, USA). This resulted in an estimated addition of 186 metric tons of total nitrogen, exceeding typical annual external nitrogen load estimates to lower Tampa Bay in a matter of days. An initial phytoplankton bloom (non-harmful diatoms) was first observed in April. Filamentous cyanobacteria blooms (*Dapis* spp.) peaked in June, followed by a bloom of the red tide organism *Karenia brevis*. Reported fish kills tracked *K. brevis* concentrations, prompting cleanup of over 1600 metric tons of dead fish. Seagrasses had minimal changes over the study period. By comparing these results to baseline environmental monitoring data, we demonstrate adverse water quality changes in response to abnormally high and rapidly delivered nitrogen loads.

**Key words:** eutrophication, macroalgae, nitrogen, phosphate mining, seagrass, Tampa Bay

## Introduction

Wastewater byproducts from mining are a global threat to the quality of surface and groundwater resources (Hudson-Edwards et al., 2011; Tayibi et al., 2009). The production of phosphate fertilizer generates large amounts of phosphogypsum waste ( $\text{CaSO}_4 \cdot \text{H}_2\text{O}$ ) that is typically stored on-site in large earthen stacks (gypstacks) capable of holding hundreds of millions of liters of process water. Water quality in gypstacks can vary depending on processing method used at the mining facility, background geological characteristics of the region, and on-site practices for managing stormwater or other activities that can introduce additional materials to the holding ponds (Henderson, 2004; Pérez-López et al., 2010). In addition to elevated phosphorus concentrations, other nutrients, contaminants, and radionuclides may be present at values much higher than natural surface waters (Beck et al., 2018a; Burnett and Elzerman, 2001). Many of these gypstacks no longer support active mining and aging infrastructure combined with climate change and seasonal stressors (e.g., heavy precipitation events) have reduced the capacity of these facilities to maintain water on site. Numerous studies have documented the environmental and human health risks associated with these stacks (Beck et al., 2018a; El Zrelli et al., 2015; Pérez-López et al., 2016; Sanders et al., 2013; Tayibi et al., 2009).

The geology of central Florida is rich in phosphates that have supported a multi-billion dollar mining industry for fertilizer to support agricultural production (Henderson, 2004). By 2001, an estimated 36 million metric tons of phosphogypsum were created each year in northern and central Florida (Burnett and Elzerman, 2001). Effective management and final closure of these facilities are imperative to reduce threats to prior ecosystem recovery efforts and investments.

The Piney Point facility located in Palmetto, Florida is a large, remnant gypstack with three

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4 holding ponds located 3 kilometers from the shore of Tampa Bay and near two Florida Aquatic  
5 Preserves [see supplement for a history of the facility; [Henderson \(2004\)](#)]. Holding capacity of  
6 the ponds has decreased over time from seasonal rain events, tropical storms, and storage of  
7 dredging material from nearby Port Manatee. Releases from the stacks occurred in the early  
8 2000s and in 2011 to nearby Bishop Harbor connected to Tampa Bay. Those releases resulted in  
9 spatially-restricted, ecosystem responses including localized harmful algal blooms and increased  
10 macroalgal abundance ([Garrett et al., 2011](#); [Switzer et al., 2011](#)).  
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13 In March 2021, leakages were detected from a tear in the plastic liner of the southern holding  
14 pond (NGS-S) at Piney Point. At that time, approximately 1.8 billion liters of mixed legacy  
15 phosphate mining wastewater and seawater from port dredging operations were being held in the  
16 failing gypstack. Piney Point historically produced Diammonium Phosphate ((NH<sub>4</sub>)<sub>2</sub>HPO<sub>4</sub>) and  
17 the remnant stackwater has very high concentrations of nitrogen, in addition to phosphorus.  
18 Water quality parameters of NGS-S measured in 2019 showed total phosphorus (160 mg/L) and  
19 total nitrogen (230 mg/L) were approximately three orders of magnitude higher than typical  
20 concentrations in Tampa Bay. From March 30th to April 9th, approximately 814 million liters  
21 (215 million gallons) of stack water were released to lower Tampa Bay following an [emergency](#)  
22 [order](#) authorized by the Florida Department of Environmental Protection (FDEP). Over this ten  
23 day period, an estimated 186 metric tons (205 tons) of nitrogen were delivered to the bay,  
24 exceeding contemporary annual estimates of external nutrient loads to lower Tampa Bay in a  
25 matter of days ([Janicki Environmental, Inc., 2017](#)).  
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28 This paper provides an initial assessment of environmental conditions in Tampa Bay over the six  
29 month period after the release of legacy phosphate mining wastewater from the Piney Point  
30 facility in 2021. The goal is to describe the results of monitoring data of surface waters collected  
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in response to the event to assess relative deviation of current conditions from long-term, seasonal records of water quality, phytoplankton, and seagrass/macroalgae datasets available for the region. Numerous studies, as well as the successful nutrient management paradigm, have demonstrated nitrogen-limitation in Tampa Bay and the system is generally considered phosphorus enriched (Greening et al., 2014; Greening and Janicki, 2006; Wang et al., 1999). As such, we focus on nitrogen in our analyses as the identified limiting nutrient for Tampa Bay and its potential to create water quality conditions unfavorable for seagrass growth due to enhanced algal production. Our analysis evaluated datasets that are descriptive of the vulnerability of seagrasses to nutrient pollution though cascading negative effects of nitrogen, phytoplankton growth and persistence, and water clarity on seagrass growth and survival (Beck et al., 2018b; Dixon and Leverone, 1995; Greening and Janicki, 2006; Kenworthy and Fonseca, 1996). A timeline of events is provided, which is supported by the quantitative results from 2021 routine and response-based monitoring of conditions in and around Port Manatee, FL – the focal point of emergency releases from the Piney Point facility. The results from this paper provide an unprecedented chronology of short-term estuarine response to acute nutrient loadings from legacy mining facilities, where context would not have been possible without the long-term monitoring datasets available for the region.

## Methods

### Simulation modeling

Monitoring of the natural resources of Tampa Bay in response to the release from Piney Point began in April, 2021 and continued for six months through September. These data were collected

through a coordinated effort under the guidance of a plume simulation by a numerical circulation model run by the Ocean Circulation Lab at the University of South Florida (USF), College of Marine Science. The plume evolution from Piney Point was simulated using the Tampa Bay Coastal Ocean Model (TBCOM) nowcast/forecast system (Chen et al., 2019, 2018), with an embedded tracer module that included realistic release rates. Normalized tracer distributions were automatically updated each day, providing 1-day hindcasts and 3.5-day forecasts throughout the period of discharge and subsequent Tampa Bay distribution. The modeled plume evolution web product (<http://ocgweb.marine.usf.edu/~liu/Tracer/>) served as the principal guidance for coordinating the data collection during the event. Preliminary model results for Piney Point are reported in Liu et al. (2021) and previous model veracity testing was described in Chen et al. (2018) and Chen et al. (2019) (and references therein).

## **Monitoring response to the emergency release**

Monitoring agencies and local partners that collected data using standardized protocols included FDEP, Environmental Protection Commission (EPC) of Hillsborough County, Parks and Natural Resources Department of Manatee County, Pinellas County Division of Environmental Management, Fish and Wildlife Research Institute (FWRI) of the Florida Fish and Wildlife Conservation Commission (FWC), City of St. Petersburg, Tampa Bay Estuary Program (TBEP), Sarasota Bay Estuary Program, Environmental Science Associates, University of South Florida, University of Florida, and New College of Florida. Monitoring efforts focused on a suite of parameters expected to respond to increased nutrient loads into the bay, including water quality sampling, phytoplankton identification, and seagrass and macroalgae transect surveys (Figure 1).

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4 Water quality parameters included discrete, laboratory-processed and *in situ* samples for total  
5 nitrogen (mg/L), total ammonia nitrogen ( $\text{NH}_3 + \text{NH}_4^+$ , mg/L, hereafter referred to as ammonia),  
6 nitrate/nitrite ( $\text{NO}_3^- + \text{NO}_2^-$ , mg/L), total phosphorus (mg/L), orthophosphate ( $\text{PO}_4^{3-}$ , mg/L),  
7 chlorophyll-a ( $\mu\text{g}/\text{L}$ ), pH, salinity (ppt), temperature ( $^{\circ}\text{C}$ ), and dissolved oxygen saturation (%).  
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10 Most samples were surface collections by boat, with sample frequency approximately biweekly  
11 for locations around Piney Point, although effort varied by monitoring group and was more  
12 consistent during the first three months after the release. Established laboratory and field sample  
13 protocols for all survey methods were based on an [Interagency Monitoring Project Plan](#)  
14 maintained by the TBEP and those of the inter-agency partners. Data quality objectives followed  
15 guidelines outlined in the USEPA-approved TBEP Data Quality Management Plan ([Sherwood et](#)  
16 [al., 2020](#)). Many of the local partners also participate in the Southwest Florida [Regional Ambient](#)  
17 [Monitoring Program](#) (RAMP) that ensures similar standards and protocols are followed in the  
18 collection and processing of monitoring data, including routine cross-reference of split samples  
19 between laboratories to check precision of measured values. Samples requiring laboratory  
20 analysis (e.g., nutrient assays) were obtained primarily from bottle collection at the surface,  
21 whereas *in situ* measurements were available for many parameters (e.g., dissolved oxygen,  
22 Secchi depth, etc.). *In situ* measurements were collected using common monitoring equipment,  
23 such as YSI sondes or Seabird CTD casts, depending on monitoring agency. Laboratory methods  
24 used to process samples were based on accepted procedures promoted through the Southwest  
25 Florida RAMP.

26 Phytoplankton samples included a mix of quantitative (cells/L) and qualitative  
27 (presence/absence) samples for major taxa at similar frequency and spatial distribution as the  
28 water quality samples. Harmful Algal Bloom (HAB) data for *Karenia brevis* were obtained from  
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4 event-based monitoring samples from the FWC-FWRI HAB Monitoring Database. HAB  
5 sampling typically occurs in response to bloom events or fish kills with extensive quality control  
6 of cell counts conducted by FWC-FWRI (additional details in Stumpf et al., 2022). HAB data  
7 were restricted to Tampa Bay boundaries and over 90% of the samples were collected within one  
8 meter of the surface. Bloom sizes for *K. brevis* were described qualitatively as low/medium/high  
9 concentrations based on FWC breakpoints at 10,000/100,000/1,000,000 cells/L. Fish kill reports  
10 were obtained from the FWC online database. Seagrass and macroalgae sampling occurred  
11 approximately biweekly at 38 transects using a modified rapid assessment design, where species  
12 were identified and enumerated using Braun-Blanquet abundances in a 0.25 m<sup>2</sup> quadrat at 10m  
13 distances along each 50m transect (see supplement). Finally, precipitation and wind data were  
14 from Albert Whitted Airfield at St. Petersburg, Florida and inflow estimates to Tampa Bay were  
15 based on summed hydrologic loads of major tributaries from US Geological Survey gaged sites  
16 (similar to Janicki Environmental, Inc., 2012). Additional details of the sampling methods and  
17 data sources are provided in supplement.

## 39 Data analysis

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41 Long-term water quality monitoring data from Hillsborough and Manatee counties (accessible at  
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43 <https://wateratlas.usf.edu/>, Hillsborough County collected monthly, Manatee County collected  
44 quarterly) were used to establish baseline conditions for major areas of interest in Figure 1a to  
45 compare with the response monitoring data described above. These areas (Area 1: closest to  
46 Piney Point; Area 2: north of Piney Point; Area 3: south of Piney Point including northern  
47 Sarasota Bay) were identified based on anticipated impacts from expected plume patterns  
48 following the TBCOM simulations and other prominent bay boundaries relative to Piney Point  
49 (i.e., the main shipping channel in the bay, inflow boundaries, location of the Skyway Bridge at  
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the mouth of Tampa Bay, and major bay segments used by TBEP for assessing annual water quality targets). Observations at each long-term monitoring station were averaged for each month across years from 2006 to 2020. This period represents a “recovery” stage for Tampa Bay where water quality conditions were much improved from historical conditions during a more eutrophic period and when seagrass areal coverage was trending towards and above a 1950s benchmark target of 15,378 hectares (38,000 acres, [Greening et al., 2014](#); [Sherwood et al., 2017](#)). For each month, the mean values +/- 1 standard deviation for each parameter at each station were quantified and used as reference values relative to results at the closest water quality monitoring station that was sampled in response to Piney Point. This comparison was made to ensure that the response data were evaluated relative to stations that were spatially relevant (e.g., long-term conditions near the mouth of Tampa Bay are not the same as those in the middle of the bay) and seasonally-specific (e.g., historical conditions in April are not the same as historical conditions in July). In some cases, the nearest long-term station did not include data for every monitoring parameter at a response location and the next closest station was used as a reference. The average distance from a monitoring location in 2021 to the long-term sites was 1.6 km (see <https://shiny.tbep.org/piney-point/> for a map of the matches).

The historical monitoring data were also used to model an expected seasonal pattern for water quality parameters from April to October in 2021. This was done by estimating smoothed annual and seasonal splines with Generalized Additive Models (GAMs) using data only from the “recovery” stage of Tampa Bay (2006 to 2020). GAMs were used to model time series of water quality parameters as a function of a continuous value for year (i.e., decimal year) and as an integer value for day of year. The continuous year value was modeled with a thin plate regression spline and the day of year value was modeled with a cyclic spline (following similar

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4 methods as Murphy et al., 2019). The modeled results provided an estimate of the expected  
5 normal seasonal variation that takes into account a long-term annual trend. Differences in the  
6 observed values sampled in the April to October time periods from the “forecasted” predictions  
7 of the baseline GAMs through 2021 provided an assessment of how the current data may have  
8 deviated from historical and normal seasonal variation.  
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15 Statistical assessments were conducted only on total nitrogen (TN), chlorophyll-a (chl-a), and  
16 Secchi disk depth as a general analysis of potential patterns in eutrophication in nitrogen-limited  
17 systems. Spatial comparisons were based primarily on the three areas identified in Figure 1a.  
18 Variables with log-normal distributions were  $\log_{10}$ -transformed (i.e., nutrients, chl-a) prior to  
19 analysis. Only the water quality data from FDEP were used for statistical analysis given the  
20 consistency of sample location and collection dates. Secchi observations that were visually  
21 identified on the bottom (71 of 431 observations in the FDEP data) were removed from analysis.  
22 Observations for other parameters that were below laboratory standards of detection were  
23 evaluated with methods described below.  
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40 Differences in observations between months for April to September for water quality, seagrass,  
41 and macroalgae within each area (Figure 1a) were evaluated using a Kruskal-Wallis one-way  
42 analysis of variance (ANOVA) followed by multiple comparisons using 2-sided Mann-Whitney  
43 U tests (Hollander et al., 2013). These tests were used to statistically characterize the temporal  
44 progression of changes in the bay following release from Piney Point, e.g., were July conditions  
45 significantly different from April? Probability values were adjusted using the sequential  
46 Bonferroni method described in (Holm, 1979) to account for the increased probability of Type I  
47 error rates with multiple comparisons. An adjusted p-value  $< 5\%$  ( $\alpha = 0.05$ ) was considered a  
48 significant difference between months. For water quality variables, monthly averages from long-  
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term monitoring data were subtracted from 2021 observations to account for normal seasonal variation not attributed to potential effects from Piney Point. Similar corrections were not done for monthly comparisons of seagrass and macroalgae data because comparable long-term seasonal data do not exist. Frequency occurrence estimates were used to evaluate macroalgae and seagrasses as a standard metric used in previous analyses in Tampa Bay (Johansson, 2016; Sherwood et al., 2017). Methods used to accommodate measured concentrations of water quality variables that were below detection included summary statistics (e.g., median, mean, and standard deviation) following estimates of the empirical cumulative distribution functions for each parameter using the Kaplan-Meier method for censored data (Helsel, 2005; Lee, 2020).

The R statistical programming language (v4.0.2) was used for all analyses (R Core Team, 2021). We imported data using the googlesheets4 (Bryan, 2020) and googledrive (D'Agostino McGowan and Bryan, 2020) R packages and used tidyverse (Wickham et al., 2019) packages to format data for analysis. The tbeptools R package (Beck et al., 2021b) was used to import and summarize long-term monitoring data (EPC water quality data and seagrass transect data). The NADA R package (Lee, 2020) was used for analysis of censored data. All spatial analyses were done using the simple features (sf) R package (Pebesma, 2018). The mgcv R package (Wood, 2017) was used to create the GAMs for water quality parameters. All datasets used in this study are available from an open access data archive hosted on the Knowledge Network for Biocomplexity (Beck, 2021). Materials for reproducing the analyses, figures, tables, and other content in this paper are provided in a GitHub repository. Finally, the Piney Point Environmental Monitoring Dashboard can be used to view all data included in this paper through an interactive, online application (Beck et al., 2021a). Links and details are provided in supplement.

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**Results**

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**Water quality trends**

12 Water quality conditions in the northern gypstack measured in 2019 and measured directly at the  
13 point of discharge in 2021 showed concentrations that were generally much higher for key water  
14 quality parameters as compared to baseline conditions in Tampa Bay (Table 1). Notably, total  
15 ammonia nitrogen was measured at 210 mg/L at Piney Point and in the discharge, compared to a  
16 long-term median of 0.02 mg/L in lower Tampa Bay. Similar differences for total phosphorus,  
17 TN, and chl-a were observed when comparing stack conditions with those of the ambient  
18 conditions in Tampa Bay.

19 Samples collected in the bay between April through September 2021 indicated that water quality  
20 conditions were outside of normal values expected for each month. A total of 7831 samples were  
21 collected and analyzed for chl-a, dissolved oxygen, TN, total phosphorus, total ammonia  
22 nitrogen, nitrate/nitrite, pH, salinity, Secchi depth, and temperature (Table 2). The percentage of  
23 observations outside of the normal range (mean +/- 1 standard deviation from long-term data)  
24 varied by location and parameter. For chl-a, 50% of the observations from April through  
25 September were above the normal range for Area 1 located closest to the discharge point,  
26 whereas only 6% and 22% were above for Areas 2 (to the north) and 3 (to the south),  
27 respectively. Total nitrogen concentrations were above the normal range for 37% of observations  
28 in Area 1, whereas concentrations were above for 22% of observations in Area 2 and 22% in  
29 Area 3. Secchi observations were below the normal range for 41% of observations in Area 1 and  
30 for 18% and 36% of observations in Areas 2 and 3. Notable differences were also observed for  
31 dissolved oxygen (e.g., 53% were above in Area 1, 44% in Area 2). Physical parameters  
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(salinity, temperature) and inorganic nitrogen (ammonia, nitrate/nitrite) were more often in normal ranges, although initial time series showed much higher concentrations for ammonia in April near Area 1. Ammonia concentrations near the point of discharge were observed in excess of 10 mg/L in April, about three orders of magnitude above baseline (Figures S2, S3), similar to the discharge measurements in Table 1. Inorganic nitrogen did not persist at high concentrations past April as it was likely rapidly utilized by phytoplankton (see below). Spatial variation among the parameters showed that values were generally above the normal range (or below for Secchi depth) for many locations near Piney Point (Area 1), Anna Maria Sound (Area 3), and the northern mouth of Tampa Bay (Area 3, Figure 2).

Total nitrogen, chl-a, and Secchi depth followed temporal progressions in 2021 that were distinct from long-term seasonal trends estimated from historical data (Figure 3). For Area 1, TN and chl-a concentrations were frequently above normal ranges during April. Chlorophyll-a concentrations were observed in excess of 50  $\mu\text{g}/\text{L}$ , although median concentrations for each week in April were less than 10  $\mu\text{g}/\text{L}$ . The initial chlorophyll peak was associated with a localized phytoplankton bloom generally dominated by diatoms. The initial diatom bloom did not persist past April. Chlorophyll concentrations decreased slightly until June and July when values increased again above the seasonal expectation, coincident with an increase in *K. brevis* concentrations to bloom levels. Many Secchi observations in Area 1 were lower than normal in April and July. Observations in Areas 2 and 3 were more often within the normal seasonal range, with some exceptions for TN and chl-a in Area 3 in April, May, and July.

Statistical comparisons between months for seasonally-corrected observations of TN, chl-a, and Secchi depth (Table 3) supported the results in Figure 3. Kruskal-Wallis tests that assessed if at least one of the months had significantly different observations for each parameter were

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4 significant ( $p < 0.05$ ) for TN, chl-a, and Secchi depth for Areas 1 and 3 and for TN and chl-a for  
5 Area 2 (Table 3). Further analysis with multiple comparison tests generally showed that  
6 April/May were different from June/July depending on Area and parameter, such that  
7 observations in the later months were generally higher (or lower for Secchi) corresponding to  
8 increasing *K. brevis* abundances by mid-summer.  
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## 17 Macroalgae and seagrass trends 18

19 A total of 38 transects were sampled for macroalgae and seagrass from April through September,  
20 each visited on average 1.7 times per month. Macroalgae observed along the transects varied in  
21 coverage, with red macroalgae groups having the highest frequency occurrence of 57%.  
22 Common taxa in the red group included genera *Gracilaria* and *Acanthophora*. Green macroalgae  
23 and filamentous cyanobacteria were less common, with frequency occurrences of 7% and 13%.  
24 Common taxa in the green group included genera *Ulva* and *Caulerpa*, whereas cyanobacteria  
25 biomass was dominated by the benthic filamentous genus *Dapis*. Brown macroalgae (primarily  
26 in the genus *Feldmannia*) were only observed at one transect in April (2% frequency  
27 occurrence). For seagrasses, turtle grass (*Thalassia testudinum*) was the dominant species with  
28 frequency occurrence of 50% across all locations and sample dates. Manatee grass (*Syringodium*  
29 *filiforme*) and shoal grass (*Halodule wrightii*) had similar coverage across all transects, with  
30 frequency occurrences of 31% and 33%, respectively. The frequency occurrences of seagrasses  
31 near Piney Point were similar to the long-term record of seagrass transect data available for  
32 Tampa Bay (Sherwood et al., 2017, also see <https://shiny.tbep.org/seagrasstransect-dash>), with  
33 turtle grass being the dominant species in more euhaline waters closer to the Gulf. There is no  
34 historical macroalgae record for Tampa Bay that is comparable to the spatial and temporal  
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resolution of the 2021 samples. Only annual historical data are available for seagrasses, with no seasonal data comparable to the results herein.

A typical temporal pattern for macroalgae and seagrass observed at many of the transects is shown in Figure 4, using transect S3T6 near Port Manatee as an example. Macroalgal abundances changed over the course of sampling similar to the remainder of transects sampled during the study. Red macroalgae were present in high abundances from April to May. Filamentous cyanobacteria (*Dapis* spp.) mats were first observed on May 24th and was present at all of the sample locations along this transect on June 4th and 15th. Filamentous cyanobacteria persisted through June and July, but was not observed in abundance after July 20th. Green macroalgae taxa were first observed in July, although at generally low abundances. Red macroalgae were the dominant taxa by the end of September. Overall abundance of seagrass did not change from April 22nd through September. The site is dominated by manatee grass that was observed at nearly all of the sample points along the transect at varying coverages.

Monthly summaries in frequency occurrence by area (Figure 5) provided an indication of macroalgae and seagrass trends in 2021 across all transects. No transects were sampled in Area 2 to the north of Piney Point and no transects were sampled past September in Area 1 given allocated sampling effort following projected dispersal patterns of the discharge from the TBCOM simulations. Red macroalgae was the dominant group across all months and areas, with the highest frequency occurrences observed in April (81% in Area 1, 95% in Area 3). Reductions in red macroalgae frequency occurrence were observed in June when cyanobacteria frequency occurrence peaked, with greater coverage of cyanobacteria in Area 3 (43%) compared to Area 1 (36%). Notable blooms of the filamentous cyanobacteria (*Dapis* spp.) were observed in Anna Maria Sound (Area 3) and near Port Manatee (Area 1) (Figure 1), typically observed covering

benthic and seagrass habitats, in addition to large floating mats on the surface. Green macroalgae had the second lowest frequency occurrence, although it increased slightly by the end of the study period (9% in September in Area 1, 31% in October in Area 3). For seagrass, both areas had generally stable total frequency occurrence. Turtle grass (*T. testudinum*) occurred in higher frequency occurrence in both areas (45% overall in Area 1, 58% overall in Area 3), compared to shoal grass (*H. wrightii*, 31% Area 1, 38% Area 3) and manatee grass (*S. filiforme*, 30% Area 1, 31% Area 3). Slight changes in frequency occurrence in Area 3 were observed for all species starting in July, with a slight reduction in frequency occurrence of turtle grass and an increase in shoal grass and manatee grass. Statistical analyses with multiple comparison tests confirmed the general trends described above, with significant changes observed over time only for macroalgae (Tables S1, S2). Tests using Braun Blanquet cover estimates confirmed the results from the frequency occurrence estimates (Tables S3, S4).

## Red tide impacts

On April 20th, the HAB species *Karenia brevis* was observed near Anna Maria Sound at the southern edge of the mouth of Tampa Bay. This first Tampa Bay influx likely originated from an ongoing coastal bloom in the Gulf of Mexico, as is common when red tide is observed in the bay (Flaherty and Landsberg, 2011; Steidinger and Ingle, 1972). By May 23, bloom concentrations of *K. brevis* were observed in lower Tampa Bay (lower/middle bay boundary Figure 1b), with concentrations peaking ( $10^6$  to  $10^7$  cells/L) by the week of July 4th in middle Tampa Bay, after which concentrations declined (Figure 6b). The increase in *K. brevis* from April to July was an anomaly in 2021 that is not regularly observed in Tampa Bay. The historical record from 1953 to present (Figure 6a) shows cell concentrations sampled in Tampa Bay between April and September, with only a few years having cell concentrations greater than  $10^5$  cells/L, notably

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4 1963, 1971, 2005, 2018, and 2021. Median cell concentrations for most years were well below  
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6 1,000 cells/L. The two highest concentrations in the long-term record were observed in 1971 (20  
7 million cells/L) and 2021 (17.6 million cells/L), both being over an order of magnitude above the  
8 high category. Cumulative rainfall and associated inflow from the main rivers entering Tampa  
9 Bay in 2021 were below historical values (2006 - 2020) in the months preceding the highest  
10 bloom concentrations (i.e., January to June, Figure 6c, d). This likely contributed to elevated  
11 salinity in lower and middle Tampa Bay that created conditions favorable for *K. brevis* growth in  
12 2021 (Figure S2f, S3f), in addition to the elevated nutrient concentrations from the Piney Point  
13 discharge.  
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16 Fish kill reports attributed to *K. brevis* at the cities of Tampa and Saint Petersburg, FL closely  
17 tracked cell concentrations during June and July 2021 (Figure 6e). In total, 331 reports were  
18 made in Saint Petersburg and 65 in Tampa. The combined weekly reports in 2021 for Tampa and  
19 Saint Petersburg peaked the week of July 4th, the same week as the peak of *K. brevis* cell  
20 concentrations (Figure 6b). Notably, all of the fish kill reports occurred within a 1.5 month  
21 period when *K. brevis* cell concentrations were consistently above the medium threshold ( $10^4$   
22 cells/L). The center of Tropical Storm Elsa (Figure 6f, pre-, post-storm wind roses) also passed  
23 through the bay area on July 5th, causing a shift in winds that likely disturbed the water column  
24 and altered the spatial distribution of *K. brevis* in the bay. Strong southeasterly winds also likely  
25 moved dead fish closer to heavily populated areas of Tampa Bay, specifically near St. Petersburg  
26 and Tampa, contributing to an increase in fish kill reports. It is important to note that high cell  
27 concentrations ( $>10^6$  cells/L) were observed in middle Tampa Bay (Figure 6b) and fish kills  
28 were reported both before and after storm passage (Figure 6e). By August, cleanup efforts  
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4 removed over 1600 metric tons of dead fish near public and private shoreline areas (K. Hammer  
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6 Levy, Pinellas County, pers. comm. Aug. 2021).  
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## 12 Discussion

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15 The observed conditions in Tampa Bay in 2021 following releases from Piney Point provide  
16 multiples lines of evidence for an adverse environmental response to a large pulse of inorganic  
17 nitrogen into the system. Collectively, these observations show that conditions in 2021 were  
18 anomalous when compared to long-term monitoring data for Tampa Bay. These anomalous  
19 events (Figure 7) included 1) a large diatom bloom in April in the vicinity of the release at Port  
20 Manatee, 2) high abundance of filamentous cyanobacteria in Anna Maria Sound and near Port  
21 Manatee, 3) medium to high bloom concentrations of the ride tide organism *K. brevis* in lower  
22 and middle Tampa Bay from June through July, and 4) high incidence of fish kill reports  
23 prompting local governments to remove over 1600 metric tons of dead fish from shoreline areas.  
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26 The water quality conditions observed during the study period, particularly for TN, chl-a, and  
27 Secchi depth, were outside of normal seasonal ranges for many of the observations (Figures 2,  
28 Table 2). The Piney Point event also represented an anomalous volume and load of labile  
29 nitrogen released directly into lower Tampa Bay. Spill events [reported to FDEP](#) (e.g., industrial  
30 spills, service line failures, sanitary sewer overflows) provide additional context for Piney Point  
31 relative to other potential anomalous releases to Tampa Bay. An assessment of over 800 reports  
32 to FDEP for the Tampa Bay watershed over the last five years showed spill volumes for these  
33 events are small (median volume 13.7 thousand liters [TBEP unpublished analysis](#)) compared to  
34 the 814 million liters released from Piney Point. Moreover, the estimated nutrient load of 186  
35 metric tons of nitrogen to Tampa Bay from Piney Point over the ten day period, exceeded current  
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4 annual estimates of all external loading sources into lower Tampa Bay (Janicki Environmental,  
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6 Inc., 2017). External nitrogen loads to lower Tampa Bay averaged 164 metric tons per year for  
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8 the baseline period of 2006 to 2020 (<https://tbep-tech.github.io/load-estimates/>).  
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## Potential nutrient cycling

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15 The events of 2021 can be considered together to develop a narrative of the temporal shift of  
16 nutrient pools between ecosystem components of the bay from April through September, starting  
17 with the influx of inorganic nitrogen from Piney Point. Total nitrogen concentrations first peaked  
18 in April (Figure 8a), as did chl-a concentrations (Figure 8b). The initial peak in water quality  
19 parameters suggested a rapid response of the phytoplankton community as an increase in diatoms  
20 (e.g., centric species, such as *Skeletonema* sp., and also *Asterionellopsis* sp., Figure 8c) that can  
21 readily utilize inorganic forms of nitrogen that were present in the initial discharge (Bates, 1976;  
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23 Domingues et al., 2011). These results were evidenced by taxonomic enumeration of  
24 phytoplankton samples collected near Port Manatee. Water quality indicators improved slightly  
25 following the decrease in diatoms in late April, as noted by relatively lower concentrations of TN  
26 and chl-a as the bloom dispersed. However, filamentous cyanobacteria biomass increased after  
27 the initial diatom bloom and peaked in June (Figure 8d), suggesting a shift of nutrients from  
28 phytoplankton to drift macroalgae communities or changing availability of nutrient ratios  
29 creating favorable conditions for macroalgae growth (Cohen and Fong, 2006; Valiela et al.,  
30  
31 1997). During peak macroalgae growth, TN and chl-a concentrations remained relatively low as  
32 nutrients were likely retained in macroalgae, until late June and early July when *K. brevis*  
33 concentrations peaked (Figure 8e). The co-occurring decline in macroalgae and increase in *K.*  
34 *brevis* suggests a release of nutrients from the former that could have stimulated growth of the  
35 latter, although residual nutrients from the initial release from Piney Point were likely still  
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available (Liu et al., 2021). Finally, conditions were relatively stable in August and September with relatively improved water quality conditions and no dominant algal blooms.

Our quantitative results provide some evidence to support the progression of events outlined above as a flow of nutrients over time. The distinct temporal progression can be readily identified through an ordination plot (Figure S7) for the observed data in Figure 8. Weekly summaries of the data are clearly separated in the ordination into monthly groups where different communities were dominant and is partially explained by orientation of the water quality vectors relative to cyanobacteria, diatoms, and *K. brevis*. For example, total nitrogen and chlorophyll are strongly aligned with the *K. brevis* axis as nutrients were likely available in organic form during the peak of the red tide event. However, this simple analysis only demonstrates an association in the observed data and cannot be verified without additional information. Additional data to support these results could include explicit load-based estimates for all sources entering the bay through 2021 and these estimates are forthcoming. Laboratory-based methods, such as isotopic analyses of nutrient signatures found in biological tissues (e.g., macroalgae) compared to those from the release, could provide a more comprehensive description of the recycling of nitrogen from Piney Point. Additional confounding variables can also obscure the association between water quality and community changes. Bay conditions preceding the 2021 events, as well as the passage of tropical storm Elsa, could obscure these associations (described below).

## Additional interpretation of impacts

Several of the water quality responses are consistent with observations of nutrient loading in other shallow Gulf Coast estuaries (Caffrey et al., 2013; Doering et al., 2006; Greening et al., 2014). The relationship between nutrients, chl-a, and water transparency followed expectations

of reduced water quality with increased nutrient loads. Temporally, these changes were observed at different times and for different species of phytoplankton. The initial increase in chl-a was first associated with a diatom bloom in April. The red tide species *K. brevis* was also first introduced to Tampa Bay from the Gulf of Mexico in April, but was not observed at high densities in the Bay until June and July. Peaks in dissolved oxygen saturation were also observed as an indicator of elevated phytoplankton production ([Kemp and Boynton, 1980](#)), particularly in July with the peak *K. brevis* bloom (Figures S2d, S3d). Of note is that inorganic species of nitrogen, mainly ammonia, were only present at high concentrations in early April. Management concerns of the negative impacts of nutrients on water quality focused primarily on the high concentrations of ammonia in the discharge (Table 1), which can be utilized rapidly by many phytoplankton taxa ([Bates, 1976](#); [Domingues et al., 2011](#)). Low concentrations of ammonia after April may be explained by quick uptake by the initial diatom bloom, where TN that included particulate and dissolved organic sources was at high concentrations through April and again peaked in July. Variation in observed concentrations of nutrients is complex given that high concentrations may suggest availability to support phytoplankton growth, whereas low concentrations may imply cycling of available nitrogen in organic forms already utilized by different taxa, including macroalgae ([Cohen and Fong, 2006](#); [Valiela et al., 1997](#)).

Previous research for Tampa Bay has identified water quality conditions that are likely to promote seagrass growth ([Greening et al., 2014](#), and references therein; [Greening and Janicki, 2006](#)). The results observed in 2021 suggested water quality conditions were not supportive of seagrass growth, although changes were not observed and the conditions likely did not persist long enough to impact seagrasses. The long-term effects of the Piney Point discharge on the seagrass community remains uncertain. From 2018 to 2020, seagrass coverage declined by 16%

in Tampa Bay, with similar losses observed in Sarasota Bay (18%), Lemon Bay (12%), and Charlotte Harbor (23%) to the south (Southwest Florida Water Management District, unpublished results). These broader trends suggest regional drivers are affecting seagrass communities (e.g., variation in precipitation, Tomasko et al., 2020), yet local issues specific to individual bays also pose challenges to managing water quality and subtidal habitats. Recent seagrass losses in Sarasota Bay may be linked to decreased light availability from a persistent *K. brevis* bloom in 2018. Although the 2021 red tide in Tampa Bay was short-lived, potential long-term effects on seagrasses remain a concern (e.g., alteration of sediment geochemistry, Eldridge et al., 2004). Ecosystem shifts from seagrass to macroalgae dominated communities are also a concern, both in 2021 and as observed at some locations in recent years from annual transect monitoring results for Tampa Bay. In particular, increasing abundance in recent years of the green algae *Caulerpa* sp. has been observed at long-term transects that were previously dominated by seagrass. These changes may be indicative of broader ecosystem shifts concurrent with alteration of nutrient loads or system resilience at the expense of seagrass communities (Lloret et al., 2005; Stafford and Bell, 2006). Acute stressors from short-term events, such as unanticipated releases from Piney Point, create additional and often preventable challenges to managing seagrass health.

Macroalgae trends across the study period were much more dramatic than the minimal changes observed in the seagrass community. This was expected given both the documented changes from past releases from Piney Point (Switzer et al., 2011) and the more rapid response of macroalgae to changing water quality conditions relative to seagrasses (Valiela et al., 1997). In Tampa Bay, red macroalgae groups (e.g., *Gracilaria* spp., *Acanthophora* sp.) are more common than green macroalgae (e.g., *Ulva* spp., *Caulerpa* spp.) and occur earlier in the growing season.

The dominance of the red groups early in the summer followed by an increase in the green alga *Ulva* spp. may reflect a natural phenology in Tampa Bay. The most notable change in the macroalgal community in 2021 was a high abundance of filamentous cyanobacteria (i.e., *Dapis* spp.) in May and June. High abundances of *Dapis* spp. were observed in Anna Maria Sound near the mouth of Tampa Bay and near Port Manatee at the release site, which is uncommon at these locations. Long-term monitoring data describing normal seasonal variation in macroalgae are unavailable and we cannot distinguish between seasonal and interannual changes and those in potential response to the Piney Point release. Filamentous cyanobacteria has been observed during routine annual transect monitoring in Tampa Bay and it has previously been documented in public reports to the Florida Department of Environmental Protection. However, these communities can respond rapidly to external nutrient inputs ([Ahern et al., 2007](#); [Albert et al., 2005](#)), often exhibiting lagged responses with characteristic growth/decay periods similar to observations herein ([Estrella, 2013](#)), and it is not unreasonable to expect these trends to be related to nutrients from Piney Point. Although long-term seasonal data are unavailable for comparison, anecdotal reports suggested that the observed biomass in 2021 was very unusual (R. Woithe, Environmental Science Associates, pers. comm. Dec. 2021).

There were also concerns that the release from Piney may have contributed to the persistence and intensity of *K. brevis*, having negative effects on fisheries resources in June and July (Figure 6). Fisheries resources in Tampa Bay have previously been negatively affected by red tide (e.g., in 2005, [Flaherty and Landsberg, 2011](#); [Schrandt et al., 2021](#)). For past Piney Point events, [Switzer et al. \(2011\)](#) evaluated nekton communities in Bishop Harbor from November 2003 to October 2004 following discharge to this subembayment. Fish community structure and species composition did not differ compared to a pre-impact period, although HAB species

(*Prorocentrum minimum*, *Heterosigma akashiwo*), including *K. brevis* and diatoms, were observed in Bishop Harbor during this time (Garrett et al., 2011). Prior blooms in Tampa Bay were more localized and *K. brevis* was at lower abundances in comparison to the 2021 bloom event, potentially mitigating exposure of fishes to related harmful conditions. In Sarasota Bay to the south, fish activity measured by passive acoustic methods was significantly lower during a 2018 red tide event as compared to pre-bloom levels (Rycyk et al., 2020). Water quality conditions before and after passage of tropical storm Elsa may have also contributed to fish kills by reducing bottom-water dissolved oxygen. Stevens et al. (2006) documented impacts of a category 4 storm on fish resources in the Charlotte Harbor estuary, although tropical storm Elsa was much smaller and fish kills were documented prior to and after arrival of the storm. Lack of continuous monitoring data for bottom waters in Tampa Bay prevents a more detailed assessment of impacts of the storm on water quality.

Establishing causal linkages between the nutrient inputs from Piney Point and the severity of the *K. brevis* bloom observed in Tampa Bay this year is difficult in the absence of more quantitative results or mechanistic tools to support understanding. Occurrence of this species has historically been spatially distinct, with blooms originating in subsurface water offshore on the West Florida Shelf (Liu et al., 2016; Steidinger, 1975; Weisberg et al., 2019, 2014) and occasionally occurring at bloom concentrations in lower and middle Tampa Bay. Although bloom concentrations in 2021 were extreme, historical blooms have been observed in Tampa Bay with notable events occurring in 1971 (Steidinger and Ingle, 1972), 2005 (Flaherty and Landsberg, 2011), and recently in 2018 (Skripnikov et al., 2021). Seasonal persistence in Gulf waters in southwest Florida can vary between years, with some blooms lasting as short as a few weeks, while others have been present for longer than a year (the 2018 bloom lasted sixteen months, Skripnikov et

al., 2021). Severity in estuaries is typically less than Gulf waters as *K. brevis* is limited to higher salinity. Contributing factors in 2021, such as low rainfall preceding the bloom and varying wind patterns, created conditions that were favorable for growth of *K. brevis* in Tampa Bay. However, the results suggest a likely scenario that residual nutrients from the Piney Point release, or indirectly through nutrients made available from the growth and decomposition of other primary producers (e.g., diatoms, macroalgae) stimulated by inputs from Piney Point, were sufficiently available to allow growth of *K. brevis* to the concentrations observed in July (also see Medina et al., 2020). Daily simulation results from the Tampa Bay Coastal Ocean Model (Chen et al., 2019, 2018) suggested that the plume was widespread throughout the bay and persisted for many months after the release ceased at Port Manatee. Plume dispersal also suggested that both open-water and back-bay habitats were exposed to nutrient concentrations sufficient to stimulate phytoplankton production. Although Piney Point did not cause red tide (i.e., it originates in the Gulf of Mexico), the events of 2021 created conditions in Tampa Bay conducive for the extreme bloom concentrations observed in July.

In the broader context of mining impacts to surface waters, these results reinforce the understanding that legacy pollutants from phosphate mining can negatively affect environmental resources. In addition to Tampa Bay (Garrett et al., 2011; Switzer et al., 2011), other Gulf Coast estuaries have been affected by pollutants from unanticipated gypstack releases. For example, two spills have occurred in Grand Bay, Mississippi, the first in 2005 following failure of the retaining walls after a heavy rain event and the second in 2012 after passage of Hurricane Isaac when the holding capacity of the local gypstack was exceeded again with heavy rainfall (Beck et al., 2018a; Dillon et al., 2015). The historical context of Grand Bay is similar to Piney Point and other international examples, e.g., Huelva estuary in Spain (Pérez-López et al., 2016, 2010).

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4 Legacy wastewater from fertilizer production has been poorly maintained at some facilities and  
5 long-term plans are insufficient to safely dispose of remnant pollutants that pose a risk of  
6 significant impacts to coastal resources that increases over time. These are not isolated examples  
7 and enhanced regulatory oversight is needed to safely and effectively close these types of  
8 facilities (Nelson et al., 2021). Local, regional, and state partners should continue to pursue  
9 management and policy actions that can mitigate the continued threats of these facilities to the  
10 health of coastal resources. These efforts are critical to managing Gulf of Mexico ecosystems  
11 given past successes and the need to address ongoing threats of climate change, human  
12 population growth, habitat loss, severe weather events, and recurring pollutant sources.  
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## Acknowledgments

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42 The progress achieved in restoring the Tampa Bay ecosystem over recent decades would not be  
43 possible without the collaborative partnerships fostered in the region. Our partners' willingness  
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4 to adapt and implement innovative monitoring and management actions in response to Piney  
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6 Point and the ever evolving challenges threatening Tampa Bay is greatly appreciated.  
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## Figure captions

Figure 1: Areas of interest and long-term monitoring stations (a) for evaluating status and trends in response-based monitoring data and sample locations from March through September 2021 by monitoring data type (b) in response to release from Piney Point. Data types include algae sampling, seagrass and macroalgae, water quality (field-based and laboratory samples), and mixed monitoring (algae, seagrass and macroalgae, water quality). Inset shows location of Tampa Bay on the Gulf coast of Florida, USA.

Figure 2: Water quality data (raw observations) for April through September 2021 following the release from Piney Point for (a) total nitrogen (mg/L), (b) chlorophyll-a ( $\mu\text{g}/\text{L}$ ), and (c) Secchi disk depth (meters). Values outside of the normal range (above for total nitrogen and chlorophyll-a, below for Secchi) are outlined in black and those in normal range are outlined in light grey. Color ramps and point sizes show relative values (reversed for Secchi). Normal ranges are defined as within +/- 1 standard deviation of the mean for the month of observation from 2006 to 2020 for values collected at the nearest long-term monitoring site to each sample location (Figure 1a). Values below detection limits (or Secchi on bottom) are not shown.

Figure 3: Expected 2021 (a) total nitrogen (mg/L), (b) chlorophyll-a ( $\mu\text{g}/\text{L}$ ), and (c) Secchi disk depth (meters) by area based on historical seasonal models. Predictions (expected values) from the historical models for dates during and after the Piney Point release are shown in thick lines (+/- 95% confidence), with observed samples overlaid on the plots to emphasize deviation of 2021 data from historical seasonal estimates. Expected values are based on Generalized Additive Models fit to historical baseline data from 2006 to early 2021, where historical predictions are shown as thin grey lines, with darker lines for more recent years. Results are grouped by assessment areas shown in Figure 1a.

Figure 4: Results for (a) macroalgae and (b) seagrass rapid response transect surveys at a site (S3T6, -82.55866 W longitude, 27.64483 N latitude) near Piney Point. Sample dates in 2021 are shown in rows with transect meter results shown in columns (0m nearshore, 50m offshore). Results show dominance of manatee grass (*Syringodium filiforme*) and red macroalgae groups, with abundances of *Dapis* spp. (cyanobacteria) peaking in June and green macroalgae (*Ulva* spp.) increasing in July. Abundances are Braun-Blanquet coverage estimates.

Figure 5: Frequency occurrence estimates for (a) Area 1 and (b) Area 3 (see map Figure 1a for locations) for macroalgae (top) and seagrass (bottom) rapid response transect surveys across all transects ( $n = 38$ ). Estimates are grouped by sample months in 2021. Frequency occurrences are absolute for each taxon based on presence/absence, whereas the total frequency occurrence applies to any taxa observed on each transect. Points are offset slightly for readability. No transects were sampled in Area 2 to the north of Piney Point and no transects were sampled past September in Area 1 given allocated sampling effort following projected dispersal patterns of the plume from model simulations.

Figure 6: *Karenia brevis* concentrations (cells/L) (a) by year and (b) by week in 2021, (c) cumulative precipitation in 2021 compared to past years, (d) cumulative inflow in 2021 compared to past years, (e) fish kill reports in 2021, and (f) wind rose plots for 2021 with notable breaks before/after Piney Point release and tropical storm Elsa. Wind roses show relative counts of six minute observations in directional (30 degree bins, north is vertical) and speed (m/s) categories.

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4 *Figure 7: Graphical timeline of events in Tampa Bay from March 30th through September 2021*  
5 *following the release from Piney Point. Inset image shows blooms of filamentous cyanobacteria*  
6 *(*Dapis spp.*).*

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8 *Figure 8: Weekly summarized observations (medians, 2.5th to 97.5th percentiles) across all*  
9 *sampled locations for (a) total nitrogen concentrations, (b) chlorophyll-a concentrations, (c)*  
10 *diatom cell concentrations, (d) filamentous cyanobacteria abundances, and (e) *Karenia brevis**  
11 *cell concentrations. Values are summarized for all samples within each week. The values suggest*  
12 *nutrient cycling between water column phytoplankton in the initial April diatom bloom, then to*  
13 *filamentous cyanobacteria in May to June, and then to *K. brevis* peaking in early July.*

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15 Quantitative cell counts for diatoms are missing for several weeks, but see Figure S6 for  
16 frequency occurrence estimates across all dates. Diatom concentrations are based on combined  
17 cell counts from *Asterionellopsis sp.* and *Skeletonema sp.*

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## Tables

*Table 1: Measured concentrations from the phosphogypsum stack (NGS-S) at Piney Point from a 2019 sample and samples from April 2021 for relevant water quality variables. Values are compared to normal annual medians (min, max) for concentrations in lower Tampa Bay. Normal medians are based on data for a baseline period from 2006 to 2020 from long-term monitoring stations in lower Tampa Bay (Figure 1a). The 2021 samples are from the NGS-S stack on April 13th and directly from the outflow site at Port Manatee on April 6th. Missing values were not measured in the stack water or release water.*

Water quality variable	2019 stack value	2021 stack value	2021 pipe value	2006 - 2020 lower Tampa Bay median (min, max)
Nitrate/Nitrite (mg/L)	0.004	0.292	0.004	0.012 (0.007, 0.014)
NH3, NH4+ (mg/L)	210	-	210	0.019 (0.007, 0.039)
TN (mg/L)	230	-	220	0.288 (0.226, 0.385)
TP (mg/L)	160	161	140	0.082 (0.058, 0.145)
Ortho-P (mg/L)	150	155	140	0.049 (0.029, 0.055)
DO (% sat.)	107.5	-	-	90.7 (86, 92)
pH	4	-	-	8.1 (8, 8.1)
Chl-a ( $\mu$ g/L)	-	105	-	3.1 (2.3, 3.5)

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 4 *Table 2: Summary of water quality variables collected in Tampa Bay from April through*  
 5 *September 2021 following the release of water from Piney Point. Variables are grouped by*  
 6 *major areas of interest for evaluating status and trends shown in Figure 1a. Summaries are*  
 7 *median, minimum, and maximum values. Total observations (N obs.) and the percentage of*  
 8 *observations in range, above, or below normal ranges are also shown. Normal ranges are*  
 9 *defined as within +/- 1 standard deviation of the mean for the month of observation from 2006 to*  
 10 *2020 for values collected at the nearest long-term monitoring site to each sample location. The*  
 11 *final column shows the percentage of total observations that were outside of detection, defined*  
 12 *as minimum laboratory detection limits for all parameters and values on the bottom for Secchi*  
 13 *observations. Medians denoted by “-” could not be calculated due to insufficient values above*  
 14 *detection.*  
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Area	Water quality variable	Med. (Min., Max.)	N obs.	% In range	% Above	% Below	% Outside detection
1	Chl-a ( $\mu\text{g/L}$ )	4.3 (1.1, 265.01)	485	44	50	6	0
	DO (% sat.)	97.9 (28.3, 215.3)	430	30	53	17	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.005 (0, 14.86)	495	66	18	17	26
	Nitrate/Nitrite (mg/L)	0 (0, 0.14352)	517	63	19	18	70
	pH	8.1 (6.8, 9.1)	476	58	29	14	0
	Sal (ppt)	30.2 (12.9, 34.6)	441	83	4	13	0
	Secchi (m)	2.4 (0.4, 9.5)	350	37	22	41	25
	Temp (C)	25.5 (19.6, 32.9)	442	66	15	19	0
	TN (mg/L)	0.41 (0.178, 5.6)	429	59	37	4	4
	TP (mg/L)	0.12 (0.019, 3.9)	485	81	15	4	1
2	Chl-a ( $\mu\text{g/L}$ )	2.7 (1.08, 42)	78	60	6	33	0
	DO (% sat.)	95 (60.6, 153.3)	73	42	44	14	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.004 (0.002, 0.071)	76	86	1	13	21
	Nitrate/Nitrite (mg/L)	- (0.00078, 0.037)	87	63	18	18	79
	pH	8 (7.3, 8.6)	92	72	16	12	0
	Sal (ppt)	27.3 (18.1, 32.3)	73	90	0	10	0
	Secchi (m)	2 (0.5, 3.5)	44	41	41	18	39
	Temp (C)	25.3 (19.9, 31.6)	73	73	7	21	0
	TN (mg/L)	0.344 (0.068, 1.13)	63	65	22	13	14
	TP (mg/L)	0.1 (0.05, 0.235)	67	60	12	28	0
3	Chl-a ( $\mu\text{g/L}$ )	2.9 (0.93, 25.9)	254	69	22	9	0
	DO (% sat.)	98.7 (42.4, 229.9)	223	53	26	21	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.003 (0.002, 0.041)	248	55	0	45	50
	Nitrate/Nitrite (mg/L)	- (0.00078, 0.046)	267	60	9	31	89
	pH	8.1 (6.2, 9.8)	245	70	21	9	0
	Sal (ppt)	31.8 (1.4, 36.5)	294	81	8	11	0
	Secchi (m)	1.9 (0.2, 5.5)	225	46	17	36	11
	Temp (C)	27 (19.6, 32.1)	294	64	13	24	0
	TN (mg/L)	0.33 (0.152, 1.78)	249	73	22	5	10
	TP (mg/L)	0.06 (0.019, 0.589)	256	78	11	12	17

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 4 *Table 3: Comparison of total nitrogen, chlorophyll-a, and Secchi depth by areas of interest*  
 5 (*Figure 1a*) and month. Overall significance of differences of concentrations between months for  
 6 each water quality variable and area combination are shown with Chi-squared statistics based  
 7 on Kruskall-Wallis rank sum tests. Multiple comparisons with Mann-Whitney U tests (Comp.  
 8 column) were used to evaluate pairwise monthly concentrations for each water quality variable  
 9 in each area. Rows that share letters within each area and water quality variable combination  
 10 have concentrations that are not significantly different between month pairs. All statistical tests  
 11 were performed on the seasonally-corrected water quality values that were based on  
 12 observations with the long-term monthly median subtracted (observed medians are shown for  
 13 comparison). \*\*  $p < 0.005$ , \*  $p < 0.05$ , blank is not significant at  $\alpha = 0.05$ .  
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Area	Water quality variable	Chi-Sq.	Comp.	Month	N obs.	Observed median	Seasonally-corrected median	
1	TN (mg/L)	25.01**	a	Apr	135	0.390	0.008	
			b	May	32	0.360	0.110	
			ab	Jun	38	0.430	0.112	
			b	Jul	24	0.520	0.178	
			ab	Aug	25	0.470	0.065	
			ab	Sep	8	0.390	0.075	
	Chl-a ( $\mu\text{g/L}$ )		a	Apr	144	3.300	1.010	
			b	May	32	2.400	-0.870	
			a	Jun	38	6.600	1.960	
			a	Jul	24	5.600	0.310	
			c	Aug	27	3.300	-3.590	
2	Secchi (m)	47.47**	a	Apr	118	2.900	0.000	
			b	May	28	3.000	-0.600	
			b	Jun	34	2.000	-0.900	
			b	Jul	18	2.000	-0.700	
			c	Aug	15	3.500	0.400	
			c	Sep	12	3.600	0.900	
	TN (mg/L)		a	Apr	18	0.390	-0.002	
			b	May	4	0.390	0.160	
			ab	Jun	3	0.500	0.113	
			ab	Jul	3	0.510	0.097	
			ab	Aug	3	0.540	0.174	
2	Chl-a ( $\mu\text{g/L}$ )	10.76*	a	Apr	22	2.500	-1.390	
			a	May	4	2.150	-2.590	
			a	Jun	4	6.000	-1.050	
			a	Jul	3	7.200	-0.940	
			a	Aug	3	5.200	-4.940	
	Secchi (m)		3.82	a	Apr	17	2.000	
			a	May	1	2.000	0.500	
			a	Jun	3	2.100	0.700	

Area	Water quality variable	Chi-Sq.	Comp.	Month	N obs.	Observed median	Seasonally-corrected median
3	TN (mg/L)	22.13**	a	Jul	1	1.400	-0.100
			a	Apr	48	0.330	-0.010
			b	May	16	0.335	0.079
			ab	Jun	10	0.350	-0.087
			ab	Jul	12	0.365	0.043
			ab	Aug	4	0.435	0.126
Chl-a ( $\mu$ g/L)		33.62**	ab	Sep	7	0.380	0.023
			ab	Apr	48	1.900	-0.900
			ac	May	16	2.350	-0.450
			b	Jun	12	2.800	-1.580
			cd	Jul	8	4.150	0.770
			bd	Aug	4	3.200	-3.100
Secchi (m)		8.77	abcd	Sep	8	3.600	-1.500
			a	Apr	41	2.700	0.000
			a	May	16	2.200	-0.500
			a	Jun	12	2.200	-0.400
			a	Jul	12	2.200	-0.100
			a	Aug	3	2.000	-0.800
			a	Sep	11	2.200	0.000

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## Initial estuarine response to the nutrient-rich Piney Point release into Tampa Bay, Florida

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### Abstract

Legacy mining facilities pose significant risks to aquatic resources. From March 30th to April 9th, 2021, 814 million liters of ~~legacy~~ phosphate mining wastewater and marine dredge water from the Piney Point facility were released into lower Tampa Bay (Florida, USA). This resulted in an estimated addition of 186 metric tons of total nitrogen, exceeding typical annual external nitrogen load estimates to lower Tampa Bay in a matter of days. Elevated levels of An initial phytoplankton bloom (non-harmful diatoms) ~~were was~~ first observed in April ~~in the lower Bay~~. Filamentous cyanobacteria blooms (*Dapis* spp.) peaked in June, followed by a bloom of the red tide organism *Karenia brevis*. Reported fish kills tracked *K. brevis* concentrations, prompting cleanup of over 1600 metric tons of dead fish. Seagrasses had minimal changes over the study period. By comparing these results to baseline environmental monitoring data, we ~~conclude that many of the biological responses observed after the release from Piney Point are abnormal relative to historic conditions~~demonstrate adverse water quality changes in response to abnormally high and rapidly delivered nitrogen loads.

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11 *Key words:* eutrophication, macroalgae, nitrogen, phosphate mining, seagrass, Tampa Bay  
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## 15 Introduction

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17 Wastewater byproducts from mining are a global threat to the quality of surface and groundwater  
18 resources (Hudson-Edwards et al., 2011; Tayibi et al., 2009). ~~Phosphate~~ The production of  
19 ~~phosphate~~ fertilizer is produced through the “wet process” reaction to create phosphoric acid by  
20 treating mined phosphate rock with sulfuric acid (Burnett and Elzerman, 2001; Pérez López et  
21 al., 2016). ~~The process~~ generates large amounts of ~~waste, creating approximately one unit of~~  
22 ~~phosphoric acid per five units of~~ phosphogypsum waste ( $\text{CaSO}_4 \cdot \text{H}_2\text{O}$ ). ~~This waste~~ that is  
23 typically stored on-site in large earthen stacks (gypstacks) capable of holding hundreds of  
24 millions of liters of process water. Water quality in gypstacks can vary depending on processing  
25 method used at the mining facility, background geological characteristics of the region, and on-  
26 site practices for managing stormwater or other activities that can introduce additional materials  
27 to the holding ponds (Henderson, 2004; Pérez-López et al., 2010). In addition to elevated  
28 phosphorus concentrations, other nutrients, contaminants, and radionuclides may be present at  
29 values much higher than ~~those of~~ natural surface waters (Beck et al., 2018a; Burnett and  
30 Elzerman, 2001). Many of these gypstacks no longer support active mining and aging  
31 infrastructure combined with climate change and seasonal stressors (e.g., heavy precipitation  
32 events) have reduced the capacity of these facilities to maintain water on site.

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34 ~~Environmental~~ Numerous studies have documented the environmental and human health risks  
35 associated with these ~~stacks can occur through controlled or uncontrolled releases to surface~~  
36 ~~waters or groundwater contamination through leaching from unlined or poorly maintained~~ stacks  
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(Beck et al., 2018a; El Zrelli et al., 2015; Pérez-López et al., 2016; Sanders et al., 2013; Tayibi et al., 2009).

Ongoing threats and challenges to protecting water quality of northern Gulf of Mexico estuaries persist despite recent environmental recovery. Successful restoration has been accomplished through ecosystem management paradigms that are based primarily on the control of nutrient pollutants from atmospheric deposition, stormwater, and wastewater. Nitrogen inputs from external sources are well understood as drivers of algal blooms in coastal environments that can degrade water quality, having a negative effect on inter- and subtidal habitats (Greening et al., 2014; Howarth and Marino, 2006; Nixon, 1995; Parker et al., 2012). Seagrasses in particular are valuable habitat-defining species that provide many ecosystem services, but are especially vulnerable to the impacts of nutrient pollution on water quality because of cascading negative effects of nitrogen, phytoplankton growth and persistence, water clarity, light limitation, and epiphyte loading on seagrass growth and survival (Beck et al., 2018b; Dixon and Leverone, 1995; Greening and Janicki, 2006; Kenworthy and Fonseca, 1996). Historical gains in seagrass coverage in southwest Florida estuaries have been achieved through public-private partnerships and consensus-based approaches to science applications that seek to limit the total nutrient loads delivered to surface waters (Greening et al., 2016; Janicki and Wade, 1996; Tomasko et al., 2020, 2018). Together, these efforts have resulted in the long-term recovery of Tampa Bay and other Gulf Coast estuaries through a reduction in external nitrogen loads, improvements in water clarity, and baywide expansion of seagrass coverage to benchmark targets established for the region (Greening et al., 2014; Sherwood et al., 2017). The three contiguous National Estuary Programs of southwest Florida (Tampa Bay, Sarasota Bay, Charlotte Harbor) and their partners

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11 have been instrumental in coordinating efforts to address legacy pollutants and current threats to  
12 the long term protection of estuarine resources.  
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15 The geology of central Florida is rich in phosphates that have supported a multi-billion dollar  
16 mining industry for fertilizer to support agricultural production ~~in other countries~~ (Henderson,  
17 2004). By 2001, an estimated 36 million metric tons of phosphogypsum were created each year  
18 in northern and central Florida (Burnett and Elzerman, 2001). ~~Currently, seventeen~~  
19 ~~phosphogypsum stacks exist in the Tampa Bay watershed.~~ Effective management and final  
20 closure of these facilities are imperative to reduce threats to prior ecosystem recovery efforts and  
21 investments. The Piney Point facility located in Palmetto, Florida is a large, remnant gypstack  
22 with three holding ponds located 3 kilometers from the shore of Tampa Bay and near two Florida  
23 Aquatic Preserves [see supplement for a history of the facility; Henderson (2004)]. Holding  
24 capacity of the ponds has decreased over time from seasonal rain events, tropical storms, and  
25 storage of dredging material from nearby Port Manatee. ~~Unanticipated releases~~Releases from the  
26 stacks occurred in the early 2000s and in 2011 to nearby Bishop Harbor connected to Tampa  
27 Bay. Those releases resulted in spatially-restricted, ecosystem responses including localized  
28 harmful algal blooms and increased macroalgal abundance (Garrett et al., 2011; Switzer et al.,  
29 2011).  
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32 ~~More recently~~In March 2021, leakages were detected from a tear in the plastic liner of the  
33 southern holding pond (NGS-S) at Piney Point. ~~In response, the Florida Department of~~  
34 ~~Environmental Protection (FDEP) authorized an emergency order on March 29th, 2021 to~~  
35 ~~release water from the southern gypstack directly into lower Tampa Bay to prevent catastrophic~~  
36 ~~failure of the facility.~~ At that time, approximately 1.8 billion liters of mixed legacy phosphate  
37 mining wastewater and seawater from port dredging operations were being held in the failing  
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11 gypstack. Piney Point historically produced Diammonium Phosphate ((NH<sub>4</sub>)<sub>2</sub>HPO<sub>4</sub>) and the  
12 remnant stackwater has very high concentrations of ~~both nitrogen, in addition to~~ phosphorus ~~and~~  
13 ~~nitrogen~~. Water quality parameters of NGS-S measured in 2019 showed total phosphorus (160  
14 mg/L) and total nitrogen (230 mg/L) ~~were approximately three orders~~  
15 ~~of magnitude higher than~~ typical ~~of surface waters~~ concentrations in Tampa Bay. From March  
16 30th to April 9th, approximately 814 million liters (215 million gallons) of stack water were  
17 released to lower Tampa Bay ~~- following an emergency order authorized by the Florida~~  
18 ~~Department of Environmental Protection (FDEP)~~. Over this ten day period, an estimated 186  
19 metric tons (205 tons) of nitrogen were delivered to the bay, exceeding contemporary annual  
20 estimates of external nutrient loads to lower Tampa Bay in a matter of days ([Janicki](#)  
21 [Environmental, Inc., 2017](#)).

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23 This paper provides an initial assessment of environmental conditions in Tampa Bay over the six  
24 month period after the release of legacy phosphate mining wastewater from the Piney Point  
25 facility in 2021. The goal is to describe the results of monitoring data of surface waters collected  
26 in response to the event to assess relative deviation of current conditions from long-term,  
27 seasonal records of water quality, phytoplankton, and seagrass/macroalgae datasets available for  
28 the region. ~~We focus on nitrogen inputs~~ Numerous studies, as well as the successful nutrient  
29 management paradigm, have demonstrated nitrogen-limitation in Tampa Bay and the system is  
30 generally considered phosphorus enriched ([Greening et al., 2014; Greening and Janicki, 2006;](#)  
31 [Wang et al., 1999](#)). As such, we focus on nitrogen in our analyses as the identified limiting  
32 nutrient for Tampa Bay and its potential to create water quality conditions unfavorable for  
33 seagrass growth due to enhanced algal production. ~~Our analysis evaluated datasets that are~~  
34 ~~descriptive of the vulnerability of seagrasses to nutrient pollution though cascading negative~~

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11 effects of nitrogen, phytoplankton growth and persistence, and water clarity on seagrass growth  
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13 and survival (Beck et al., 2018b; Dixon and Leverone, 1995; Greening and Janicki, 2006;  
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15 Kenworthy and Fonseca, 1996). A timeline of events is provided, which is supported by the  
16 quantitative results from 2021 routine and response-based monitoring of conditions in and  
17 around Port Manatee, FL – the focal point of emergency releases from the Piney Point facility.  
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19 The results from this paper provide examples an unprecedented chronology of anticipated short-  
20 term responses estuarine response to acute nutrient loadings from legacy mining facilities in the  
21 broader, where context of historical conditions that may influence system response to these  
22 events would not have been possible without the long-term monitoring datasets available for the  
23 region.

## 31 Methods

### 32 Simulation modeling

### 33 Monitoring response to the emergency release

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35 Monitoring of the natural resources of Tampa Bay in response to the release from Piney Point  
36 began in April, 2021 and continued for six months through September. These data were collected  
37 through a coordinated effort under the guidance of a plume simulation by a numerical circulation  
38 model run by the Ocean Circulation Lab at the University of South Florida (USF), College of  
39 Marine Science. The plume evolution from Piney Point was simulated using the Tampa Bay  
40 Coastal Ocean Model (TBCOM) nowcast/forecast system (Chen et al., 2019, 2018), with an  
41 embedded tracer module that included realistic release rates. Normalized tracer distributions  
42 were automatically updated each day, providing 1-day hindcasts and 3.5-day forecasts  
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10 throughout the period of discharge and subsequent Tampa Bay distribution. The modeled plume  
11 evolution web product ([Y. Liu, R.H. Weisberg, J. Chen, Y. Sun, University of South Florida, pers. comm. Apr. 2021](#)) (<http://ocgweb.marine.usf.edu/~liu/Tracer/>) served as the principal  
12 guidance for coordinating the data collection during the event. [Preliminary model results for](#)  
13 [Piney Point are reported in Liu et al. \(2021\) and previous model veracity testing was described in](#)  
14 [Chen et al. \(2018\) and Chen et al. \(2019\) \(and references therein\).](#)

### **Monitoring response to the emergency release**

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22 Monitoring agencies and local partners that collected data using standardized protocols included  
23 FDEP, Environmental Protection Commission (EPC) of Hillsborough County, Parks and Natural  
24 Resources Department of Manatee County, Pinellas County Division of Environmental  
25 Management, Fish and Wildlife Research Institute (FWRI) of the Florida Fish and Wildlife  
26 Conservation Commission (FWC), City of St. Petersburg, Tampa Bay Estuary Program (TBEP),  
27 Sarasota Bay Estuary Program, Environmental Science Associates, University of South Florida,  
28 University of Florida, and New College of Florida. Monitoring efforts focused on a suite of  
29 parameters expected to respond to increased nutrient loads into the bay, including water quality  
30 sampling, phytoplankton identification, and seagrass and macroalgae transect surveys (Figure 1).

#### **In short, water**

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32 [Water](#) quality parameters included discrete, laboratory-processed and *in situ* samples for total  
33 nitrogen (mg/L), total ammonia nitrogen ( $\text{NH}_3 + \text{NH}_4^+$ , mg/L, [hereafter referred to as ammonia](#)),  
34 nitrate/nitrite ( $\text{NO}_3^- + \text{NO}_2^-$ , mg/L), total phosphorus (mg/L), orthophosphate ( $\text{PO}_4^{3-}$ , mg/L),  
35 chlorophyll-a ( $\mu\text{g}/\text{L}$ ), pH, salinity (ppt), temperature ( $^{\circ}\text{C}$ ), and dissolved oxygen saturation (%).  
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37 Most samples were surface collections by boat, with sample frequency approximately biweekly

for locations around Piney Point, although effort varied by monitoring group and was more consistent during the first three months after the release. Established laboratory and field sample protocols for all survey methods were based on an Interagency Monitoring Project Plan maintained by the TBEP and those of the inter-agency partners. Data quality objectives followed guidelines outlined in the USEPA-approved TBEP Data Quality Management Plan (Sherwood et al., 2020). Many of the local partners also participate in the Southwest Florida Regional Ambient Monitoring Program (RAMP) that ensures similar standards and protocols are followed in the collection and processing of monitoring data, including routine cross-reference of split samples between laboratories to check precision of measured values. Samples requiring laboratory analysis (e.g., nutrient assays) were obtained primarily from bottle collection at the surface, whereas in situ measurements were available for many parameters (e.g., dissolved oxygen, Secchi depth, etc.). In situ measurements were collected using common monitoring equipment, such as YSI sondes or Seabird CTD casts, depending on monitoring agency. Laboratory methods used to process samples were based on accepted procedures promoted through the Southwest Florida RAMP.

Phytoplankton samples included a mix of quantitative (cells/L) and qualitative (presence/absence) samples for major taxa at similar frequency and spatial distribution as the water quality samples. Harmful Algal Bloom (HAB) data for *Karenia brevis* were obtained from event-based monitoring samples from the FWC-FWRI HAB Monitoring Database.

Phytoplankton bloomsHAB sampling typically occurs in response to bloom events or fish kills with extensive quality control of cell counts conducted by FWC-FWRI (additional details in Stumpf et al., 2022). HAB data were restricted to Tampa Bay boundaries and over 90% of the samples were collected within one meter of the surface. Bloom sizes for *K. brevis* were described

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11 qualitatively as low/medium/high concentrations based on FWC breakpoints at  
12 10,000/100,000/1,000,000 cells/L. Fish kill reports were obtained from the FWC online  
13 database. Seagrass and macroalgae sampling occurred approximately biweekly at 38 transects  
14 using a modified rapid assessment design ~~following the “Eyes on Seagrass” method developed~~  
15 ~~by a local citizen science group in cooperation with academic, where species were identified~~ and  
16 ~~federal partners enumerated using Braun-Blanquet abundances in a 0.25 m<sup>2</sup> quadrat at 10m~~  
17 ~~distances along each 50m transect (see supplement).~~ Finally, precipitation and wind data were  
18 ~~from Tampa International Airport, Albert Whitted Airfield at St. Petersburg, Florida and~~ inflow  
19 estimates to Tampa Bay were ~~based~~based on summed hydrologic loads of major tributaries from  
20 US Geological Survey gaged sites, ~~and wind data were from Albert Whitted Airfield at~~  
21 ~~St. Petersburg, Florida (similar to Janicki Environmental, Inc., 2012).~~ Additional details of the  
22 sampling methods and data sources are provided in supplement.  
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### 34 **Data Analysis**

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36 Long-term water quality monitoring data from Hillsborough and Manatee counties (accessible at  
37 <https://wateratlas.usf.edu/>, Hillsborough County collected monthly, Manatee County collected  
38 quarterly) were used to establish baseline conditions for major areas of interest in Figure 1a to  
39 compare with the response monitoring data described above. These areas (Area 1: closest to  
40 Piney Point; Area 2: north of Piney Point; Area 3: south of Piney Point including northern  
41 Sarasota Bay) were identified based on anticipated impacts from expected plume patterns  
42 following the TBCOM simulations and other prominent bay boundaries relative to Piney Point  
43 (i.e., the main shipping channel in the bay, inflow boundaries, location of the Skyway Bridge at  
44 the mouth of Tampa Bay, and major bay segments used by TBEP for assessing annual water  
45 quality targets). Observations at each long-term monitoring station were averaged for each  
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month across years from 2006 to 2020. This period represents a “recovery” stage for Tampa Bay where water quality conditions were much improved from historical conditions during a more eutrophic period and when seagrass areal coverage was trending towards and above a 1950s benchmark target of 15,378 hectares (38,000 acres, [Greening et al., 2014](#); [Sherwood et al., 2017](#)). For each month, the mean values +/- 1 standard deviation for each parameter at each station were quantified and used as reference values relative to results at the closest water quality monitoring station that was sampled in response to Piney Point. This comparison was made to ensure that the response data were evaluated relative to stations that were spatially relevant (e.g., long-term conditions near the mouth of Tampa Bay are not the same as those in the middle of the bay) and seasonally-specific (e.g., historical conditions in April are not the same as historical conditions in July). In some cases, the nearest long-term station did not include data for every monitoring parameter at a response location and the next closest station was used as a reference. The average distance from a monitoring location in 2021 to the long-term sites was 1.6 km (see <https://shiny.tbep.org/piney-point/> for a map of the matches).

The historical monitoring data were also used to model an expected seasonal pattern for water quality parameters from April to October in 2021. This was done by estimating smoothed annual and seasonal splines with Generalized Additive Models (GAMs) using data only from the “recovery” stage of Tampa Bay (2006 to 2020). GAMs were used to model time series of water quality parameters as a function of a continuous value for year (i.e., decimal year) and as an integer value for day of year. The continuous year value was modeled with a thin plate regression spline and the day of year value was ~~modelled~~modeled with a cyclic spline (following similar methods as [Murphy et al., 2019](#)). The modeled results provided an estimate of the expected normal seasonal variation that takes into account a long-term annual trend. Differences

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11 in the observed values sampled in the April to October time periods from the “forecasted”  
12 predictions of the baseline GAMs through 2021 provided an assessment of how the current data  
13 may have deviated from historical and normal seasonal variation.  
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15 Statistical assessments were conducted only on total nitrogen (TN), chlorophyll-a (chl-a), and  
16 Secchi disk depth as a general analysis of potential patterns in eutrophication in nitrogen-limited  
17 systems. ~~Observations for each data type were typically aggregated to the weekly or monthly~~  
18 ~~scale given that sampling occurred on different days during the six month period.~~ Spatial  
19 comparisons were based primarily on the three areas identified in Figure 1a. Variables with log-  
20 normal distributions were  $\log_{10}$ -transformed (i.e., nutrients, chl-a) prior to analysis. ~~For~~  
21 ~~statistical tests using Only the~~ water quality data, ~~only the monitoring results~~ from FDEP were  
22 used for statistical analysis given the consistency of sample location and collection dates. Secchi  
23 observations that were visually identified on the bottom (71 of 431 observations in the FDEP  
24 data) were removed from analysis. Observations for other parameters that were below laboratory  
25 standards of detection were evaluated with methods described below.  
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27  
28 Differences in observations between months for April to September for water quality, seagrass,  
29 and macroalgae within each area (Figure 1a) were evaluated using a Kruskal-Wallis one-way  
30 analysis of variance (ANOVA) followed by multiple comparisons using 2-sided Mann-Whitney  
31 U tests (Hollander et al., 2013). These tests were used to statistically characterize the temporal  
32 progression of changes in the bay following release from Piney Point, e.g., were July conditions  
33 significantly different from April? Probability values were adjusted using the sequential  
34 Bonferroni method described in (Holm, 1979) to account for the increased probability of Type I  
35 error rates with multiple comparisons. An adjusted p-value < 5% ( $\alpha = 0.05$ ) was considered a  
36 significant difference between months. For water quality variables, monthly averages from long-  
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term monitoring data were subtracted from 2021 observations to account for normal seasonal variation not attributed to potential effects from Piney Point. Similar corrections were not done for monthly comparisons of seagrass and macroalgae data because comparable long-term seasonal data do not exist. [Frequency occurrence estimates were used to evaluate macroalgae and seagrasses as a standard metric used in previous analyses in Tampa Bay \(Johansson, 2016; Sherwood et al., 2017\)](#). Methods used to accommodate measured concentrations of water quality variables that were below detection included summary statistics (e.g., median, mean, and standard deviation) following estimates of the empirical cumulative distribution functions for each parameter using the Kaplan-Meier method for censored data ([Helsel, 2005](#); [Lee, 2020](#)).

The R statistical programming language (v4.0.2) was used for all analyses ([R Core Team, 2021](#)). We imported data using the googlesheets4 ([Bryan, 2020](#)) and googledrive ([D'Agostino McGowan and Bryan, 2020](#)) R packages and used tidyverse ([Wickham et al., 2019](#)) packages to format data for analysis. The tbeptools R package ([Beck et al., 2021b](#)) was used to import and summarize long-term monitoring data (EPC water quality data and seagrass transect data). The NADA R package ([Lee, 2020](#)) was used for analysis of censored data. All spatial analyses were done using the simple features (sf) R package ([Pebesma, 2018](#)). The mgcv R package ([Wood, 2017](#)) was used to create the GAMs for water quality parameters. All datasets used in this study are available from an open access data archive hosted on the Knowledge Network for Biocomplexity ([Beck, 2021](#)). Materials for reproducing the analyses, figures, tables, and other content in this paper are provided in a [GitHub repository](#). Finally, the Piney Point Environmental Monitoring Dashboard can be used to view all data included in this paper through an interactive, online application ([Beck et al., 2021a](#)). Links and details are provided in supplement.

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11 **Results**  
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14 **Timeline of 2021 biological response events**  
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17 A general summary of 2021 events in Tampa Bay following discharge from Piney Point is  
18 shown in Figure 2. After the discharge ceased on April 9th, a peak in median chl a concentration  
19 was observed near Piney Point (Area 1, Figure 1a) in mid April, with peak individual sample  
20 values in excess of 50  $\mu\text{g/L}$ . Median concentrations for each week in April were less than 10  
21  $\mu\text{g/L}$ . The discharge phytoplankton assemblage was comprised of over 99% of a spherical  
22 nanoplanktonic chlorophyte ( $3.37 \times 10^8$  cells/L). The phytoplankton communities near the  
23 discharge area in April were generally dominated by diatoms. The initial diatom bloom did not  
24 persist past April. On April 20th, the HAB species *Karenia brevis* was observed near Anna  
25 Maria Sound at the southern edge of the mouth of Tampa Bay; this first Tampa Bay influx was  
26 related to an ongoing coastal bloom. By May/June, bloom levels of *K. brevis* were observed in  
27 lower Tampa Bay (lower/middle bay boundary Figure 1b), with peak concentrations in excess of  
28  $1 \times 10^6$  cells/L. Also during May/June, high abundances of filamentous cyanobacteria (*Davis*  
29 spp.) were observed in Anna Maria Sound (Area 3) and near Port Manatee (Area 1). High levels  
30 of cyanobacteria coverage on benthic and seagrass habitats were observed, in addition to large  
31 floating mats on the surface. By June 27th, fish kill reports attributed to *K. brevis* increased as  
32 cellular abundance climbed in lower and middle Tampa Bay. The center of tropical storm Elsa  
33 passed to the west of Tampa Bay on July 5th, causing a shift in prevailing winds from the  
34 southeast. This shift in winds likely disturbed the water column and altered the spatial  
35 distribution of *K. brevis* in the bay. Strong southeasterly winds also likely moved dead fish closer  
36 to heavily populated areas of Tampa Bay, specifically near the cities of St. Petersburg and  
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11 Tampa, contributing to an increase in fish kill reports. Concentrations of *K. brevis* in middle and  
12 lower Tampa Bay peaked in early to mid July, with bloom conditions not observed in the bay  
13 after July. Conditions were relatively stable in August and September compared to months prior.  
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15 A quantitative description of these events follows.

## 16 Water quality trends

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19 Water quality conditions in the northern gypstack measured in 2019 and measured directly at the  
20 point of discharge in 2021 showed concentrations that were generally much higher for key water  
21 quality parameters as compared to baseline conditions in Tampa Bay (Table 1). Notably, total  
22 ammonia nitrogen was measured at 210 mg/L at Piney Point and in the discharge, compared to a  
23 long-term median of 0.02 mg/L in lower Tampa Bay. Similar differences for total phosphorus,  
24 TN, and chl-a were observed when comparing stack conditions with those of the ambient  
25 conditions in Tampa Bay. These contrasts provided a context for the monitoring data collected in  
26 2021.

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28 Samples collected in the bay between April through September 2021 indicated that water quality  
29 conditions were unusual and outside of normal values expected for each month. A total of 7831  
30 samples were collected and analyzed for chl-a, dissolved oxygen, TN, total phosphorus, total  
31 ammonia nitrogen, nitrate/nitrite, pH, salinity, Secchi depth, and temperature (Table 2). The  
32 percentage of observations outside of the normal range (mean +/- 1 standard deviation from  
33 long-term data) varied by location and parameter. For chl-a, 50% of the observations from April  
34 through September were above the normal range for Area 1 located closest to the discharge  
35 point, whereas only 6% and 22% were above for Areas 2 (to the north) and 3 (to the south),  
36 respectively. Total nitrogen concentrations were above the normal range for 37% of observations

in Area 1, whereas concentrations were above for 22% of observations in Area 2 and 22% in Area 3. Secchi observations were below the normal range for 41% of observations in Area 1 and for 18% and 36% of observations in Areas 2 and 3. Notable differences were also observed for dissolved oxygen (e.g., 53% were above in Area 1, 44% in Area 2). Physical parameters (salinity, temperature) and inorganic nitrogen (ammonia, nitrate/nitrite) were more often in normal ranges, although initial time series showed much higher concentrations for ammonia in April near Area 1. Ammonia concentrations near the point of discharge were observed in excess of 10 mg/L in April, about three orders of magnitude above baseline (Figures S2, S3), similar to the discharge measurements in Table 1~~1~~. Inorganic nitrogen did not persist at high concentrations past April as it was likely rapidly utilized by phytoplankton (see below). Spatial variation among the parameters showed that values were generally above the normal range (or below for Secchi depth) for many locations near Piney Point (Area 1), Anna Maria Sound (Area 3), and the northern mouth of Tampa Bay (Area 3, Figure 32).

Total nitrogen, chl-a, and Secchi depth followed temporal progressions in 2021 that were distinct from long-term seasonal trends estimated from historical data (Figure 43). For Area 1, TN and chl-a concentrations were frequently above normal ranges during April. Chlorophyll-a concentrations were observed in excess of 50 µg/L, although median concentrations for each week in April were less than 10 µg/L. The initial chlorophyll peak was associated with a localized phytoplankton bloom generally dominated by diatoms. The initial diatom bloom did not persist past April. Concentrations Chlorophyll concentrations decreased slightly until June and July when values increased again above the seasonal expectation, coincident with thean increase in *K. brevis* concentrations to bloom levels. Many Secchi observations in Area 1 were

lower than normal in April and July. Observations in Areas 2 and 3 were more often within the normal seasonal range, with some exceptions for TN and chl-a in Area 3 in April, May, and July.

Statistical comparisons between months for seasonally-corrected observations of TN, chl-a, and Secchi depth (Table 3) supported the results in Figure 43. Kruskal-Wallis tests that assessed if at least one of the months had significantly different observations for each parameter were significant ( $p < 0.05$ ) for TN, chl-a, and Secchi depth for Areas 1 and 3 and for TN and chl-a for Area 2 (Table 3). Further analysis with multiple comparison tests generally showed that April/May were different from June/July depending on Area and parameter, such that observations in the later months were generally higher (or lower for Secchi) corresponding to increasing *K. brevis* abundances by mid-summer.

### Macroalgae and seagrass trends

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A total of 38 transects were sampled for macroalgae and seagrass from April through September, each visited on average 1.7 times per month. Macroalgae observed along the transects varied in coverage, with red macroalgae groups having the highest frequency occurrence of 57%.

Common taxa in the red group included genera *Gracilaria* and *Acanthophora*. Green macroalgae and filamentous cyanobacteria were less common, with frequency occurrences of 7% and 13%.

Common taxa in the green group included genera *Ulva* and *Caulerpa*, whereas cyanobacteria biomass was dominated by the benthic filamentous genus *Dapis*. Brown macroalgae (primarily in the genus *Feldmannia*) were only observed at one transect in April (2% frequency occurrence). For seagrasses, turtle grass (*Thalassia testudinum*) was the dominant species with frequency occurrence of 50% across all locations and sample dates. Manatee grass (*Syringodium filiforme*) and shoal grass (*Halodule wrightii*) had similar coverage across all transects, with

frequency occurrences of 31% and 33%, respectively. The frequency occurrences of seagrasses near Piney Point were similar to the long-term record of seagrass transect data available for Tampa Bay (Sherwood et al., 2017, also see <https://shiny.tbep.org/seagrasstransect-dash>), with turtle grass being the dominant species in more euhaline waters closer to the Gulf. There is no historical macroalgae record for Tampa Bay that is comparable to the spatial and temporal resolution of the 2021 samples. Only annual historical data are available for seagrasses, with no seasonal data comparable to the results herein.

A typical temporal pattern for macroalgae and seagrass observed at many of the transects is shown in Figure 5. ~~Transect 4, using transect S3T6 is located less than one kilometer to the north of~~ Port Manatee as an example. Macroalgal abundances changed over the course of sampling similar to the remainder of transects sampled during the study. Red macroalgae were present in high abundances from April to May. Filamentous cyanobacteria (*Dapis* spp.) mats were first observed on May 24th and was present at all of the sample locations along this transect on June 4th and 15th. Filamentous cyanobacteria persisted through June and July, but was not observed in abundance after July 20th. Green macroalgae taxa were first observed in July, although at generally low abundances. Red macroalgae were the dominant taxa by the end of September. Overall abundance of seagrass did not change from April 22nd through September. The site is dominated by manatee grass that was observed at nearly all of the sample points along the transect at varying coverages.

Monthly summaries in frequency occurrence by area (Figure 65) provided an indication of macroalgae and seagrass trends in 2021 across all transects. No transects were sampled in Area 2 to the north of Piney Point and no transects were sampled past September in Area 1 given allocated sampling effort following projected dispersal patterns of the discharge from the

TBCOM simulations. Macroalgal dominance varied across the months, similar to the example in Figure 5. Red macroalgae was the dominant group across all months and areas, with the highest frequency occurrences observed in April (81% in Area 1, 95% in Area 3). Reductions in red macroalgae frequency occurrence were observed in June when cyanobacteria frequency occurrence peaked, with greater coverage of cyanobacteria in Area 3 (43%) compared to Area 1 (36%). Notable blooms of the filamentous cyanobacteria (*Dapis* spp.) were observed in Anna Maria Sound (Area 3) and near Port Manatee (Area 1) (Figure 1), typically observed covering benthic and seagrass habitats, in addition to large floating mats on the surface. Green macroalgae had the second lowest frequency occurrence, although it increased slightly by the end of the study period (9% in September in Area 1, 31% in October in Area 3). Brown macroalgae was only observed at one transect. For seagrass, both areas had generally stable total frequency occurrence. Turtle grass (*T. testudinum*) occurred in higher frequency occurrence in both areas (45% overall in Area 1, 58% overall in Area 3), compared to shoal grass (*H. wrightii*, 31% Area 1, 38% Area 3) and manatee grass (*S. filiforme*, 30% Area 1, 31% Area 3). Slight changes in frequency occurrence in Area 3 were observed for all species starting in July, with a slight reduction in frequency occurrence of turtle grass and an increase in shoal grass and manatee grass. Statistical analyses with multiple comparison tests confirmed the general trends described above, with significant changes observed over time only for macroalgae (Tables S1, S2). Tests using Braun Blanquet cover estimates also produced similar results from the frequency occurrence estimates (Tables S3, S4).

## Red tide impacts

Bloom concentrations of On April 20th, the red tide HAB species *K. brevis* in 2021 were first was observed near Anna Maria Sound at the southern edge of the mouth of Tampa Bay. This

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11 first Tampa Bay influx likely originated from an ongoing coastal bloom in the Gulf of Mexico,  
12 as is common when red tide is observed in Tampa Bay the week of May 23, the bay (Flaherty and  
13 Landsberg, 2011; Steidinger and Ingle, 1972). By May 23, bloom concentrations of *K. brevis*  
14 were observed in lower Tampa Bay (lower/middle bay boundary Figure 1b), with concentrations  
15 peaking ( $10^6$  to  $10^7$  cells/L) by the week of July 4th in middle Tampa Bay, after which  
16 concentrations declined (Figure 7b6b). The increase in *K. brevis* from April to July was an  
17 anomaly in 2021 that is not regularly observed in Tampa Bay. The historical record from 1953 to  
18 present (Figure 7a6a) shows cell concentrations sampled in Tampa Bay between April and  
19 September, with only a few years having cell concentrations greater than  $10^5$  cells/L, notably  
20 1963, 1971, 2005, 2018, and 2021. Median cell concentrations for most years were well below  
21 1,000 cells/L. The two highest concentrations in the long-term record were observed in 1971 (20  
22 million cells/L) and 2021 (17.6 million cells/L), both being over an order of magnitude above the  
23 high category. Cumulative rainfall and associated inflow from the main rivers entering Tampa  
24 Bay in 2021 were below historical values (1995–2006 – 2020) in the months preceding the highest  
25 bloom concentrations (i.e., January to June, Figure 7e6c, d). This likely contributed to elevated  
26 salinity in lower and middle Tampa Bay that created conditions favorable for *K. brevis* growth in  
27 2021 (Figure S2f, S3f), in addition to the elevated nutrient concentrations from the Piney Point  
28 discharge.  
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30 Fish kill reports attributed to *K. brevis* at the cities of Tampa and Saint Petersburg, FL closely  
31 tracked cell concentrations during June and July 2021 (Figure 7e6e). In total, 331 reports were  
32 made in Saint Petersburg and 65 in Tampa. The combined weekly reports in 2021 for Tampa and  
33 Saint Petersburg peaked the week of July 4th, the same week as the peak of *K. brevis* cell  
34 concentrations (Figure 7b6b). Notably, all of the fish kill reports occurred within a 1.5 month  
35 period.  
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10 period when *K. brevis* cell concentrations were consistently above the medium threshold ( $10^4$  cells/L). The center of Tropical Storm Elsa (Figure 7f6f, pre-, post-storm wind roses) also passed through the bay area on July 5th, causing a shift in winds that likely ~~altered the location of *K. brevis* cells and dead fish in the bay, disturbed the water column and altered the spatial distribution of *K. brevis* in the bay. Strong southeasterly winds also likely moved dead fish closer to heavily populated areas of Tampa Bay, specifically near St. Petersburg and Tampa, contributing to an increase in fish kill reports.~~ It is important to note that high cell concentrations ( $>10^6$  cells/L) were observed in middle Tampa Bay (Figure 7b6b) and fish kills were reported both before and after storm passage (Figure 7e6e). By August, ~~Pinellas County and the city of St. Petersburg cleanup efforts~~ removed over 1600 metric tons of dead fish near public and private shoreline areas (K. Hammer Levy, Pinellas County, pers. comm. Aug. 2021).

### 32 Potential nutrient cycling

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35 The above results can be considered together to develop a narrative of the temporal shift of  
36 nutrient pools between ecosystem components of the bay from April through September, starting  
37 with the influx of inorganic nitrogen from Piney Point. Total nitrogen concentrations first peaked  
38 in April (Figure 8a), as did chl a concentrations (Figure 8b). The initial peak in water quality  
39 parameters suggested a rapid response of the phytoplankton community as an increase in diatoms  
40 (e.g., centric species, such as *Skeletonema* sp., and also *Asterionellopsis* sp., Figure 8c) that can  
41 readily utilize inorganic forms of nitrogen that were present in the initial discharge (Bates, 1976;  
42 Domingues et al., 2011). Water quality indicators improved slightly following the decrease in  
43 diatoms in late April, as noted by relatively lower concentrations of TN and chl a. However,  
44 filamentous cyanobacteria biomass increased after the initial diatom bloom and peaked in June  
45 (Figure 8d), suggesting a shift of nutrients from phytoplankton to drift macroalgae communities.  
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11 During peak macroalgae growth, TN and chl-a concentrations remained relatively low as  
12 nutrients were likely retained in macroalgae, until late June and early July when *K. brevis*  
13 concentrations peaked (Figure 8e). The co occurring decline in macroalgae and increase in *K.*  
14 *brevis* suggests a release of nutrients from the former that could have stimulated growth of the  
15 latter, although residual water column nutrients from Piney Point may have also been present (as  
16 suggested by modelling efforts). Finally, conditions were relatively stable in August and  
17 September with relatively improved water quality conditions and no dominant algal blooms.  
18 These distinct temporal periods were readily identified through an ordination plot (Figure S7) for  
19 the observed data in Figure 8.  
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## 29 Discussion

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31 The observed conditions in Tampa Bay in 2021 following unanticipated releases from Piney  
32 Point provide multiples lines of evidence for an adverse environmental response to a large pulse  
33 of inorganic nitrogen into the system. Collectively, these observations show that conditions in  
34 2021 were anomalous when compared to long-term monitoring data for Tampa Bay. These  
35 anomalous events (Figure 7) included 1) a large diatom bloom in April in the vicinity of the  
36 release at Port Manatee, 2) high abundance of filamentous cyanobacteria in Anna Maria Sound  
37 and near Port Manatee, 3) medium to high bloom concentrations of the ride tide organism *K.*  
38 *brevis* in lower and middle Tampa Bay from June through July, and 4) high incidence of fish kill  
39 reports prompting local governments to remove over 1600 metric tons of dead fish from  
40 shoreline areas. The water quality conditions observed during the study period, particularly for  
41 TN, chl-a, and Secchi depth, were outside of normal seasonal ranges for many of the  
42 observations (Figures 32, Table 2). The Piney Point event also represented an anomalous volume  
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11 and load of labile nitrogen released directly into lower Tampa Bay. Spill events reported to  
12 FDEP (e.g., industrial spills, service line failures, sanitary sewer overflows) provide additional  
13 context for Piney Point relative to other potential anomalous releases to Tampa Bay. An  
14 assessment of over 800 reports to FDEP for the Tampa Bay watershed over the last five years  
15 showed spill volumes for these events are small (median volume 13.7 thousand liters TBEP  
16 unpublished analysis) compared to the 814 million liters released from Piney Point. Moreover,  
17 the estimated nutrient load of 186 metric tons of nitrogen to Tampa Bay from Piney Point over  
18 the ten day period, exceeded current annual estimates of all external loading sources into lower  
19 Tampa Bay (Janicki Environmental, Inc., 2017). External nitrogen loads to lower Tampa Bay  
20 averaged 164 metric tons per year for the baseline period of 2006 to 2020 ([https://tbep-](https://tbep-tech.github.io/load-estimates/)  
21 [tech.github.io/load-estimates/](https://tbep-tech.github.io/load-estimates/)).

### Potential nutrient cycling

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35 The events of 2021 can be considered together to develop a narrative of the temporal shift of  
36 nutrient pools between ecosystem components of the bay from April through September, starting  
37 with the influx of inorganic nitrogen from Piney Point. Total nitrogen concentrations first peaked  
38 in April (Figure 8a), as did chl-a concentrations (Figure 8b). The initial peak in water quality  
39 parameters suggested a rapid response of the phytoplankton community as an increase in diatoms  
40 (e.g., centric species, such as *Skeletonema* sp., and also *Asterionellopsis* sp., Figure 8c) that can  
41 readily utilize inorganic forms of nitrogen that were present in the initial discharge (Bates, 1976;  
42 Domingues et al., 2011). These results were evidenced by taxonomic enumeration of  
43 phytoplankton samples collected near Port Manatee. Water quality indicators improved slightly  
44 following the decrease in diatoms in late April, as noted by relatively lower concentrations of TN  
45 and chl-a as the bloom dispersed. However, filamentous cyanobacteria biomass increased after  
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11 the initial diatom bloom and peaked in June (Figure 8d), suggesting a shift of nutrients from  
12 phytoplankton to drift macroalgae communities or changing availability of nutrient ratios  
13 creating favorable conditions for macroalgae growth (Cohen and Fong, 2006; Valiela et al.,  
14 1997). During peak macroalgae growth, TN and chl-a concentrations remained relatively low as  
15 nutrients were likely retained in macroalgae, until late June and early July when *K. brevis*  
16 concentrations peaked (Figure 8e). The co-occurring decline in macroalgae and increase in *K.*  
17 *brevis* suggests a release of nutrients from the former that could have stimulated growth of the  
18 latter, although residual nutrients from the initial release from Piney Point were likely still  
19 available (Liu et al., 2021). Finally, conditions were relatively stable in August and September  
20 with relatively improved water quality conditions and no dominant algal blooms.  
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30 Our quantitative results provide some evidence to support the progression of events outlined  
31 above as a flow of nutrients over time. The distinct temporal progression can be readily  
32 identified through an ordination plot (Figure S7) for the observed data in Figure 8. Weekly  
33 summaries of the data are clearly separated in the ordination into monthly groups where different  
34 communities were dominant and is partially explained by orientation of the water quality vectors  
35 relative to cyanobacteria, diatoms, and *K. brevis*. For example, total nitrogen and chlorophyll are  
36 strongly aligned with the *K. brevis* axis as nutrients were likely available in organic form during  
37 the peak of the red tide event. However, this simple analysis only demonstrates an association in  
38 the observed data and cannot be verified without additional information. Additional data to  
39 support these results could include explicit load-based estimates for all sources entering the bay  
40 through 2021 and these estimates are forthcoming. Laboratory-based methods, such as isotopic  
41 analyses of nutrient signatures found in biological tissues (e.g., macroalgae) compared to those  
42 from the release, could provide a more comprehensive description of the recycling of nitrogen  
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11 from Piney Point. Additional confounding variables can also obscure the association between  
12 water quality and community changes. Bay conditions preceding the 2021 events, as well as the  
13 passage of tropical storm Elsa, could obscure these associations (described below).  
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### 16 Additional interpretation of impacts

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18 Several of the water quality responses are consistent with observations of nutrient loading in  
19 other shallow Gulf Coast estuaries (Caffrey et al., 2013; Doering et al., 2006; Greening et al.,  
20 2014). The relationship between nutrients, chl-a, and water transparency followed expectations  
21 of reduced water quality with increased nutrient loads. Temporally, these changes were observed  
22 at different times and for different species of phytoplankton. The initial increase in chl-a was first  
23 associated with a diatom bloom in April. The red tide species *K. brevis* was also first introduced  
24 to Tampa Bay from the Gulf of Mexico in April, but was not observed at high densities in the  
25 Bay until June and July. Peaks in dissolved oxygen saturation were also observed as an indicator  
26 of elevated phytoplankton production (Kemp and Boynton, 1980), particularly in July with the  
27 peak *K. brevis* bloom (Figures S2, S3S2d, S3d). Of note is that inorganic species of nitrogen,  
28 mainly ammonia, were only present at high concentrations in early April. Management concerns  
29 of the negative impacts of nutrients on water quality focused primarily on the high  
30 concentrations of ammonia in the discharge (Table 1), which can be utilized rapidly by many  
31 phytoplankton taxa (Bates, 1976; Domingues et al., 2011). Low concentrations of ammonia after  
32 April may be explained by quick uptake by the initial diatom bloom, where TN that included  
33 particulate and dissolved organic sources was at high concentrations through April and again  
34 peaked in July. Variation in observed concentrations of nutrients is complex given that high  
35 concentrations may suggest availability to support phytoplankton growth, whereas low  
36 concentrations may suggest availability to support phytoplankton growth, whereas low  
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concentrations may imply cycling of available nitrogen in organic forms already utilized by different taxa, including macroalgae (Cohen and Fong, 2006; Valiela et al., 1997).

Previous research for Tampa Bay has identified water quality conditions that are likely to promote seagrass growth (Greening et al., 2014, and references therein; Greening and Janicki, 2006). The results observed in 2021 suggested water quality conditions were not supportive of seagrass growth, although changes were not observed and the conditions likely did not persist long enough to impact seagrasses. There were also concerns that the release from Piney may have contributed to the persistence and intensity of *K. brevis*, having negative effects on fisheries resources in June and July (Figure 7). In addition to fish kill reports, quantitative data on changes in nekton abundance and diversity in Tampa Bay in 2021 are forthcoming. Routine sampling of fisheries occurs monthly in Tampa Bay and a long-term record back to 1998 provides detailed information for the major bay segments. Results from the Tampa Bay Nekton Index showed a decline in fisheries resources following a significant red tide event that persisted for several months in lower Tampa Bay in 2005 (Flaherty and Landsberg, 2011; Schrandt et al., 2021). Given the observed *K. brevis* concentrations in 2021 and the magnitude of fish kills, mandates for catch and release for popular sportfishes (*Sciaenops ocellatus*, *Cynoscion nebulosus*, and *Centropomus undecimalis*) were extended through the fall of 2021. For past Piney Point events, Switzer et al. (2011) evaluated nekton communities in Bishop Harbor from November 2003 to October 2004 following discharge to this subembayment. Fish community structure and species composition did not differ compared to a pre-impact period, although HAB species (*Prorocentrum minimum*, *Heterosigma akashiwo*), including *K. brevis* and diatoms, were observed in Bishop Harbor during this time (Garrett et al., 2011). Prior blooms in Tampa Bay were more localized and *K. brevis* was at lower abundances in comparison to the 2021 bloom.

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11 event, potentially mitigating exposure of fishes to related harmful conditions. In Sarasota Bay to  
12 the south, fish activity measured by passive acoustic methods was significantly lower during a  
13 2018 red tide event as compared to pre bloom levels (Ryeik et al., 2020).  
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17 For seagrasses, major bloom events in 2021 produced unfavourable water quality conditions,  
18 although changes in frequency occurrence of seagrasses were not observed over the initial study  
19 period. The long-term effects of the Piney Point discharge on the seagrass community remains  
20 uncertain. From 2018 to 2020, seagrass coverage declined by 16% in Tampa Bay, with similar  
21 losses observed in Sarasota Bay (18%), Lemon Bay (12%), and Charlotte Harbor (23%) to the  
22 south (Southwest Florida Water Management District, unpublished results). These broader trends  
23 suggest regional drivers are affecting seagrass communities (e.g., variation in precipitation,  
24 Tomasko et al., 2020), yet local issues specific to individual bays also pose challenges to  
25 managing water quality and subtidal habitats. Recent seagrass losses in Sarasota Bay may be  
26 linked to decreased light availability from a persistent *K. brevis* bloom in 2018. Although the  
27 2021 red tide in Tampa Bay was short-lived, potential long-term effects on seagrasses remain a  
28 concern (e.g., alteration of sediment geochemistry, Eldridge et al., 2004). Ecosystem shifts from  
29 seagrass to macroalgae dominated communities are also a concern, both in 2021 and as observed  
30 at some locations in recent years from annual transect monitoring results for Tampa Bay. In  
31 particular, increasing abundance in recent years of the green algae *Caulerpa* sp. has been  
32 observed at long-term transects that were previously dominated by seagrass. These changes may  
33 be indicative of broader ecosystem shifts concurrent with alteration of nutrient loads or system  
34 resilience at the expense of seagrass communities (Lloret et al., 2005; Stafford and Bell, 2006).  
35 Acute stressors from short-term events, such as unanticipated releases from Piney Point, create  
36 additional and often preventable challenges to managing seagrass health.  
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Macroalgae trends across the study period were much more dramatic than the minimal changes observed in the seagrass community. This was expected given both the documented changes from past releases from Piney Point (Switzer et al., 2011) and the more rapid response of macroalgae to changing water quality conditions relative to seagrasses (Valiela et al., 1997). In Tampa Bay, red macroalgae groups (e.g., *Gracilaria* spp., *Acanthophora* sp.) are more common than green macroalgae (e.g., *Ulva* spp., *Caulerpa* spp.) and occur earlier in the growing season. The dominance of the red groups early in the summer followed by an increase in the green alga *Ulva* spp. may reflect a natural phenology in Tampa Bay. The most notable change in the macroalgal community in 2021 was a high abundance of filamentous cyanobacteria (i.e., *Dapis* spp.) in May and June. High abundances of *Dapis* spp. were observed in Anna Maria Sound near the mouth of Tampa Bay and near Port Manatee at the release site, which is uncommon at these locations. Long-term monitoring data describing normal seasonal variation in macroalgae are unavailable and we cannot distinguish between seasonal and interannual changes and those in potential response to the Piney Point release. Filamentous cyanobacteria has been observed during routine annual transect monitoring in Tampa Bay and it has previously been documented in public reports to the Florida Department of Environmental Protection. However, these communities can respond rapidly to external nutrient inputs (Ahern et al., 2007; Albert et al., 2005), often exhibiting lagged responses with characteristic growth/decay periods similar to observations herein (Estrella, 2013), and it is not unreasonable to expect these trends to be related to nutrients from Piney Point. Although long-term seasonal data are unavailable for comparison, anecdotal reports suggested that the observed biomass in 2021 was very unusual (R. Woithe, Environmental Science Associates, pers. comm. Dec. 2021).

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11 There were also concerns that the release from Piney may have contributed to the persistence and  
12 intensity of *K. brevis*, having negative effects on fisheries resources in June and July (Figure 6).  
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14 Fisheries resources in Tampa Bay have previously been negatively affected by red tide (e.g., in  
15 2005, Flaherty and Landsberg, 2011; Schrandt et al., 2021). For past Piney Point events, Switzer  
16 et al. (2011) evaluated nekton communities in Bishop Harbor from November 2003 to October  
17 2004 following discharge to this subembayment. Fish community structure and species  
18 composition did not differ compared to a pre-impact period, although HAB species  
19 (*Prorocentrum minimum*, *Heterosigma akashiwo*), including *K. brevis* and diatoms, were  
20 observed in Bishop Harbor during this time (Garrett et al., 2011). Prior blooms in Tampa Bay  
21 were more localized and *K. brevis* was at lower abundances in comparison to the 2021 bloom  
22 event, potentially mitigating exposure of fishes to related harmful conditions. In Sarasota Bay to  
23 the south, fish activity measured by passive acoustic methods was significantly lower during a  
24 2018 red tide event as compared to pre-bloom levels (Rycyk et al., 2020). Water quality  
25 conditions before and after passage of tropical storm Elsa may have also contributed to fish kills  
26 by reducing bottom-water dissolved oxygen. Stevens et al. (2006) documented impacts of a  
27 category 4 storm on fish resources in the Charlotte Harbor estuary, although tropical storm Elsa  
28 was much smaller and fish kills were documented prior to and after arrival of the storm. Lack of  
29 continuous monitoring data for bottom waters in Tampa Bay prevents a more detailed  
30 assessment of impacts of the storm on water quality.  
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10 Shelf (Liu et al., 2016; Steidinger, 1975; Weisberg et al., 2019, 2014) and occasionally occurring  
11 at bloom concentrations in lower and middle Tampa Bay. Although bloom concentrations in  
12 2021 were extreme, historical blooms have been observed in Tampa Bay with notable events  
13 occurring in 1971 ([Steidinger and Ingle, 1972](#)[Steidinger and Ingle, 1972](#)), 2005 (Flaherty and  
14 Landsberg, 2011), and recently in 2018 (Skripnikov et al., 2021). [Seasonal persistence in Gulf](#)  
15 [waters in southwest Florida can vary between years, with some blooms lasting as short as a few](#)  
16 [weeks, while others have been present for longer than a year \(the 2018 bloom lasted sixteen](#)  
17 [months, Skripnikov et al., 2021\).](#) Severity in estuaries is typically less than Gulf waters as *K.*  
18 *brevis* is limited to higher salinity. Contributing factors in 2021, such as low rainfall preceding  
19 the bloom and varying wind patterns, ~~also~~-created conditions that were favorable for growth of  
20 *K. brevis* [in Tampa Bay](#). However, the results suggest a likely scenario that residual nutrients  
21 from the Piney Point release, or indirectly through nutrients made available from the growth and  
22 decomposition of other primary producers (e.g., diatoms, macroalgae) stimulated by inputs from  
23 Piney Point, were sufficiently available to allow growth of *K. brevis* to the concentrations  
24 observed in July (also see Medina et al., 2020). Daily simulation results from the Tampa Bay  
25 Coastal Ocean Model (Chen et al., 2019, 2018) suggested that the plume was widespread  
26 throughout the bay and persisted for many months after the release ceased at Port Manatee.  
27 Plume dispersal also suggested that both open-water and back-bay habitats were exposed to  
28 nutrient concentrations sufficient to stimulate phytoplankton production. Although Piney Point  
29 did not cause red tide (i.e., it originates in the Gulf of Mexico), the events of 2021 created  
30 conditions in Tampa Bay conducive for the extreme bloom concentrations observed in July.  
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32 Additional monitoring and analysis are also required to fully understand the long-term impacts to  
33 bay resources beyond water quality. For benthic communities, sediments sampled in April and  
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September near Port Manatee and surrounding waters suggested a mix of conditions dominated by “intermediate” and “healthy” benthic index scores (Wade et al., 2006; see [https://shiny.tbep.org/piney\\_point](https://shiny.tbep.org/piney_point)), possibly reflecting the generally high spatial variability of macroinvertebrate communities in coastal habitats (Gillett et al., 2021; Karlen et al., 2020). Comparison of the April and September samples to historical conditions suggested relatively consistent benthic invertebrate community structure from 1993 to present (TBEP unpublished results). Differences between the April and September samples were not observed. Finally, effects of changing environmental conditions and *K. brevis* on marine mammals (e.g., cetaceans, sirenians) was also a concern given their use of bay resources. Twenty manatee (*Trichechus manatus latirostris*) mortalities were reported in the red tide boundary of the impacted counties of Tampa Bay through August 2021. This is of particular concern given the recent unusual mortality events for Florida manatees that are likely linked to seagrass losses on the east coast of Florida (e.g., Indian River Lagoon) and current seagrass losses for southwest Florida.

In the broader context of mining impacts to surface waters, these results reinforce the understanding that legacy pollutants from phosphate mining can negatively affect environmental resources. In addition to Tampa Bay (Garrett et al., 2011; Switzer et al., 2011), other Gulf Coast estuaries have been affected by pollutants from unanticipated gypstack releases. Grand Bay is a 7500 hectare protected area in southern Mississippi that has been exposed to phosphorus rich and highly acidic water from a defunct gypstack (Beck et al., 2018a; Dillon et al., 2015). Two spills have occurred in Grand Bay, Mississippi (Dillon et al., 2015). Two spills have occurred in Grand Bay, the first in 2005 following failure of the retaining walls after a heavy rain event and the second in 2012 after passage of Hurricane Isaac when the holding capacity of the local gypstack was exceeded again with heavy rainfall (Beck et al., 2018a; Dillon et al., 2015). gypstack was

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11 exceeded again with heavy rainfall. Massive fish kills were observed and likely related to low pH  
12 of the water released. Unlike Piney Point, inorganic nitrogen concentrations of the release were  
13 low due to a different fertilizer production method and concerns of the long-term impacts  
14 focused primarily on heavy loads of orthophosphate (Dillon et al., 2015). Phosphate loads to  
15 Tampa Bay from Piney Point were similar in magnitude to the nitrogen loads, although  
16 concentrations were within normal baseline ranges within a month after the release stopped  
17 (Figures S2, S3). The fate of the phosphorus in Tampa Bay is less understood than that of  
18 nitrogen. Regardless, the historical context of Grand Bay is similar to Piney Point and other  
19 international examples, e.g., Huelva estuary in Spain (Pérez-López et al., 2016, 2010). Legacy  
20 wastewater from fertilizer production has been poorly maintained at some facilities and long-  
21 term plans are insufficient to safely dispose of remnant pollutants that pose a risk of significant  
22 impacts to coastal resources that increases over time. These are not isolated examples and  
23 enhanced regulatory oversight is needed to safely and effectively close these types of facilities  
24 (Nelson et al., 2021).

37 Limitations of our analyses are also important to note to inform future event-based monitoring  
38 and additional research. All of the analyses are correlative based on associations between the  
39 measured water quality observations, macroalgae, and seagrass results and may not represent  
40 explicit cause-and-effect mechanisms. However, the interpretations are supported by previous  
41 research on drivers of primary production and eutrophication of coastal waters. Additional data  
42 to support these results could include explicit load-based estimates for all sources entering the  
43 bay through 2021 and these estimates are forthcoming. Laboratory-based methods, such as  
44 isotopic analyses of nutrient signatures found in biological tissues (e.g., macroalgae) compared  
45 to those from the release, could provide a more comprehensive description of the recycling of  
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11 nitrogen from Piney Point. Long term fate of nutrients from Piney Point is uncertain and  
12 continued monitoring can further support understanding of ecosystem response in Tampa Bay  
13 beyond the initial results in 2021. Local, regional, and state partners should continue to pursue  
14 management and policy actions that can mitigate the continued threats of these facilities to the  
15 health of coastal resources. These efforts are critical to managing Gulf of Mexico ecosystems  
16 given past successes and the need to address ongoing threats of climate change, human  
17 population growth, habitat loss, severe weather events, and recurring pollutant sources.  
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## Acknowledgments

26  
27 We thank the many TBEP partners and collaborators for their continuing efforts to restore and  
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29 Florida Department of Environmental Protection, Environmental Protection Commission of  
30 Hillsborough County, Parks and Natural Resources Department of Manatee County, Pinellas  
31 County Division of Environmental Management, Fish and Wildlife Research Institute of the  
32 Florida Fish and Wildlife Conservation Commission, City of St. Petersburg, [Tampa Bay Watch](#),  
33 Sarasota Bay Estuary Program, Environmental Science Associates, University of South Florida,  
34 University of Florida, and New College of Florida. Funding was provided to the University of  
35 Florida from the Ocean Conservancy and the National Science Foundation (project ID 2130675).  
36 The progress achieved in restoring the Tampa Bay ecosystem over recent decades would not be  
37 possible without the collaborative partnerships fostered in the region. Our partners' willingness  
38 to adapt and implement innovative monitoring and management actions in response to Piney  
39 Point and the ever evolving challenges threatening Tampa Bay is greatly appreciated.  
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*Figure 1: Areas of interest and long-term monitoring stations (a) for evaluating status and trends in response-based monitoring data and sample locations from March through September 2021 by monitoring data type (b) in response to release from Piney Point. Data types include algae sampling, seagrass and macroalgae, water quality (field-based and laboratory samples), and mixed monitoring (algae, seagrass and macroalgae, water quality). Inset shows location of Tampa Bay on the Gulf coast of Florida, USA.*

*Figure 2*~~Figure 2~~ Graphical timeline of events in Tampa Bay from March 30th through September 2021 following the release from Piney Point. Inset image shows blooms of filamentous cyanobacteria (*Dapis* spp.).

*Figure 3:* Water quality data (raw observations) for April through September 2021 following the release from Piney Point for (a) total nitrogen (mg/L), (b) chlorophyll-a (µg/L), and (c) Secchi disk depth (meters). Values outside of the normal range (above for total nitrogen and chlorophyll-a, below for Secchi) are outlined in black and those in normal range are outlined in light grey. Color ramps and point sizes show relative values (reversed for Secchi). Normal ranges are defined as within +/- 1 standard deviation of the mean for the month of observation from 2006 to 2020 for values collected at the nearest long-term monitoring site to each sample location (Figure 1a). Values below detection limits (or Secchi on bottom) are not shown.

*Figure 4*~~3~~: Expected 2021 (a) total nitrogen (mg/L), (b) chlorophyll-a (µg/L), and (c) Secchi disk depth (meters) by area based on historical seasonal models. Predictions (expected values) from the historical models for dates during and after the Piney Point release are shown in thick lines (+/- 95% confidence), with observed samples overlaid on the plots to emphasize deviation of 2021 data from historical seasonal estimates. Expected values are based on Generalized Additive Models fit to historical baseline data from 2006 to early 2021, where historical predictions are shown as thin grey lines, with darker lines for more recent years. Results are grouped by assessment areas shown in Figure 1a.

*Figure 5*~~4~~: Results for (a) macroalgae and (b) seagrass rapid response transect surveys at a site (S3T6, -82.55866 W longitude, 27.64483 N latitude) near Piney Point. Sample dates in 2021 are shown in rows with transect meter results shown in columns (0m nearshore, 50m offshore). Results show dominance of manatee grass (*Syringodium filiforme*) and red macroalgae groups, with abundances of *Dapis* spp. (cyanobacteria) peaking in June and green macroalgae (*Ulva* spp.) increasing in July. Abundances are Braun-Blanquet coverage estimates.

*Figure 6*~~5~~: Frequency occurrence estimates for (a) Area 1 and (b) Area 3 (see map Figure 1a for locations) for macroalgae (top) and seagrass (bottom) rapid response transect surveys across all transects ( $n = 38$ ). Estimates are grouped by sample months in 2021. Frequency occurrences are absolute for each taxon based on presence/absence, whereas the total frequency occurrence applies to any taxa observed on each transect. Points are offset slightly for readability. No transects were sampled in Area 2 to the north of Piney Point and no transects were sampled past September in Area 1 given allocated sampling effort following projected dispersal patterns of the plume from model simulations.

*Figure 7*~~6~~: *Karenia brevis* concentrations (cells/L) (a) by year and (b) by week in 2021, (c) cumulative precipitation in 2021 compared to past years, (d) cumulative inflow in 2021 compared to past years, (e) fish kill reports in 2021, and (f) wind rose plots for 2021 with

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11 notable breaks before/after Piney Point release and tropical storm Elsa. Wind roses show  
12 relative counts of six minute observations in directional (30 degree bins, north is vertical) and  
13 speed (m/s) categories.

14 Figure 7: Graphical timeline of events in Tampa Bay from March 30th through September 2021  
15 following the release from Piney Point. Inset image shows blooms of filamentous cyanobacteria  
16 (*Dapis spp.*)

17 Figure 8: Weekly summarized observations (medians, 2.5th to 97.5th percentiles) across all  
18 sampled locations for (a) total nitrogen concentrations, (b) chlorophyll-a concentrations, (c)  
19 diatom cell concentrations, (d) filamentous cyanobacteria abundances, and (e) *Karenia brevis*  
20 cell concentrations. Values are summarized for all samples within each week. The values suggest  
21 nutrient cycling between water column phytoplankton in the initial April diatom bloom, then to  
22 filamentous cyanobacteria in May to June, and then to *K. brevis* peaking in early July.  
23 Quantitative cell counts for diatoms are missing for several weeks, but see Figure S6 for  
24 frequency occurrence estimates across all dates. Diatom concentrations are based on combined  
25 cell counts from *Asterionellopsis sp.* and *Skeletonema sp.*

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## Tables

Table 1: Measured concentrations from the phosphogypsum stack (NGS-S) at Piney Point from a 2019 sample and samples from April 2021 for relevant water quality variables. Values are compared to normal annual medians (min, max) for concentrations in lower Tampa Bay. Normal medians are based on data for a baseline period from 2006 to 2020 from long-term monitoring stations in lower Tampa Bay (Figure 1a). The 2021 samples are from the NGS-S stack on April 13th and directly from the outflow site at Port Manatee on April 6th. Missing values were not measured in the stack water or release water.

Water quality variable	2019 stack value	2021 stack value	2021 pipe value	2006 - 2020 lower Tampa Bay median (min, max)
Nitrate/Nitrite (mg/L)	0.004	0.292	0.004	0.012 (0.007, 0.014)
NH3, NH4+ (mg/L)	210	-	210	0.019 (0.007, 0.039)
TN (mg/L)	230	-	220	0.288 (0.226, 0.385)
TP (mg/L)	160	161	140	0.082 (0.058, 0.145)
Ortho-P (mg/L)	150	155	140	0.049 (0.029, 0.055)
DO (% sat.)	107.5	-	-	90.7 (86, 92)
pH	4	-	-	8.1 (8, 8.1)
Chl-a ( $\mu$ g/L)	-	105	-	3.1 (2.3, 3.5)

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Table 2: Summary of water quality variables collected in Tampa Bay from April through September 2021 following the release of water from Piney Point. Variables are grouped by major areas of interest for evaluating status and trends shown in Figure 1a. Summaries are median, minimum, and maximum values. Total observations ( $N$  obs.) and the percentage of observations in range, above, or below normal ranges are also shown. Normal ranges are defined as within +/- 1 standard deviation of the mean for the month of observation from 2006 to 2020 for values collected at the nearest long-term monitoring site to each sample location. The final column shows the percentage of total observations that were outside of detection, defined as minimum laboratory detection limits for all parameters and values on the bottom for Secchi observations. Medians denoted by “-” could not be calculated due to insufficient values above detection.

Area	Water quality variable	Med. (Min., Max.)	N obs.	% In range	% Above	% Below	% Outside detection
1	Chl-a ( $\mu\text{g/L}$ )	4.3 (1.1, 265.01)	485	44	50	6	0
	DO (% sat.)	97.9 (28.3, 215.3)	430	30	53	17	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.005 (0, 14.86)	495	66	18	17	26
	Nitrate/Nitrite (mg/L)	0 (0, 0.14352)	517	63	19	18	70
	pH	8.1 (6.8, 9.1)	476	58	29	14	0
	Sal (ppt)	30.2 (12.9, 34.6)	441	83	4	13	0
	Secchi (m)	2.4 (0.4, 9.5)	350	37	22	41	25
	Temp (C)	25.5 (19.6, 32.9)	442	66	15	19	0
	TN (mg/L)	0.41 (0.178, 5.6)	429	59	37	4	4
	TP (mg/L)	0.12 (0.019, 3.9)	485	81	15	4	1
2	Chl-a ( $\mu\text{g/L}$ )	2.7 (1.08, 42)	78	60	6	33	0
	DO (% sat.)	95 (60.6, 153.3)	73	42	44	14	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.004 (0.002, 0.071)	76	86	1	13	21
	Nitrate/Nitrite (mg/L)	- (0.00078, 0.037)	87	63	18	18	79
	pH	8 (7.3, 8.6)	92	72	16	12	0
	Sal (ppt)	27.3 (18.1, 32.3)	73	90	0	10	0
	Secchi (m)	2 (0.5, 3.5)	44	41	41	18	39
	Temp (C)	25.3 (19.9, 31.6)	73	73	7	21	0
	TN (mg/L)	0.344 (0.068, 1.13)	63	65	22	13	14
	TP (mg/L)	0.1 (0.05, 0.235)	67	60	12	28	0
3	Chl-a ( $\mu\text{g/L}$ )	2.9 (0.93, 25.9)	254	69	22	9	0
	DO (% sat.)	98.7 (42.4, 229.9)	223	53	26	21	0
	NH <sub>3</sub> , NH <sub>4+</sub> (mg/L)	0.003 (0.002, 0.041)	248	55	0	45	50
	Nitrate/Nitrite (mg/L)	- (0.00078, 0.046)	267	60	9	31	89
	pH	8.1 (6.2, 9.8)	245	70	21	9	0
	Sal (ppt)	31.8 (1.4, 36.5)	294	81	8	11	0
	Secchi (m)	1.9 (0.2, 5.5)	225	46	17	36	11
	Temp (C)	27 (19.6, 32.1)	294	64	13	24	0
	TN (mg/L)	0.33 (0.152, 1.78)	249	73	22	5	10
	TP (mg/L)	0.06 (0.019, 0.589)	256	78	11	12	17

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11 **Table 3:** Comparison of total nitrogen, chlorophyll-a, and Secchi depth by areas of interest  
 12 (Figure 1a) and month. Overall significance of differences of concentrations between months for  
 13 each water quality variable and area combination are shown with Chi-squared statistics based  
 14 on Kruskall-Wallis rank sum tests. Multiple comparisons with Mann-Whitney U tests (Comp.  
 15 column) were used to evaluate pairwise monthly concentrations for each water quality variable  
 16 in each area. Rows that share letters within each area and water quality variable combination  
 17 have concentrations that are not significantly different between month pairs. All statistical tests  
 18 were performed on the seasonally-corrected water quality values that were based on  
 19 observations with the long-term monthly median subtracted (observed medians are shown for  
 20 comparison). \*\*  $p < 0.005$ , \*  $p < 0.05$ , blank is not significant at  $\alpha = 0.05$ .

Area	Water quality variable	Chi-Sq.	Comp.	Month	N obs.	Observed median	Seasonally-corrected median
1	TN (mg/L)	25.01**	a	Apr	135	0.390	0.008
			b	May	32	0.360	0.110
			ab	Jun	38	0.430	0.112
			b	Jul	24	0.520	0.178
			ab	Aug	25	0.470	0.065
			ab	Sep	8	0.390	0.075
1	Chl-a ( $\mu\text{g/L}$ )	61.84**	a	Apr	144	3.300	1.010
			b	May	32	2.400	-0.870
			a	Jun	38	6.600	1.960
			a	Jul	24	5.600	0.310
			c	Aug	27	3.300	-3.590
			a	Apr	118	2.900	0.000
1	Secchi (m)	47.47**	a	May	28	3.000	-0.600
			b	Jun	34	2.000	-0.900
			b	Jul	18	2.000	-0.700
			c	Aug	15	3.500	0.400
			c	Sep	12	3.600	0.900
			a	Apr	18	0.390	-0.002
2	TN (mg/L)	20.85**	a	May	4	0.390	0.160
			ab	Jun	3	0.500	0.113
			ab	Jul	3	0.510	0.097
			ab	Aug	3	0.540	0.174
			ab	Sep	1	0.570	0.049
			a	Apr	22	2.500	-1.390
2	Chl-a ( $\mu\text{g/L}$ )	10.76*	a	May	4	2.150	-2.590
			a	Jun	4	6.000	-1.050
			a	Jul	3	7.200	-0.940
			a	Aug	3	5.200	-4.940
			a	Apr	17	2.000	0.200
			a	May	1	2.000	0.500
2	Secchi (m)	3.82	a	Jun	3	2.100	0.700
			a	Apr	17	2.000	0.200

Area	Water quality variable	Chi-Sq.	Comp.	Month	N obs.	Observed median	Seasonally-corrected median
3	TN (mg/L)	22.13**	a	Jul	1	1.400	-0.100
			a	Apr	48	0.330	-0.010
			b	May	16	0.335	0.079
			ab	Jun	10	0.350	-0.087
			ab	Jul	12	0.365	0.043
			ab	Aug	4	0.435	0.126
			ab	Sep	7	0.380	0.023
	Chl-a ( $\mu\text{g/L}$ )	33.62**	ab	Apr	48	1.900	-0.900
			ab	May	16	2.350	-0.450
			b	Jun	12	2.800	-1.580
			cd	Jul	8	4.150	0.770
			bd	Aug	4	3.200	-3.100
			abcd	Sep	8	3.600	-1.500
	Secchi (m)	8.77	a	Apr	41	2.700	0.000
			a	May	16	2.200	-0.500
			a	Jun	12	2.200	-0.400
			a	Jul	12	2.200	-0.100
			a	Aug	3	2.000	-0.800
			a	Sep	11	2.200	0.000

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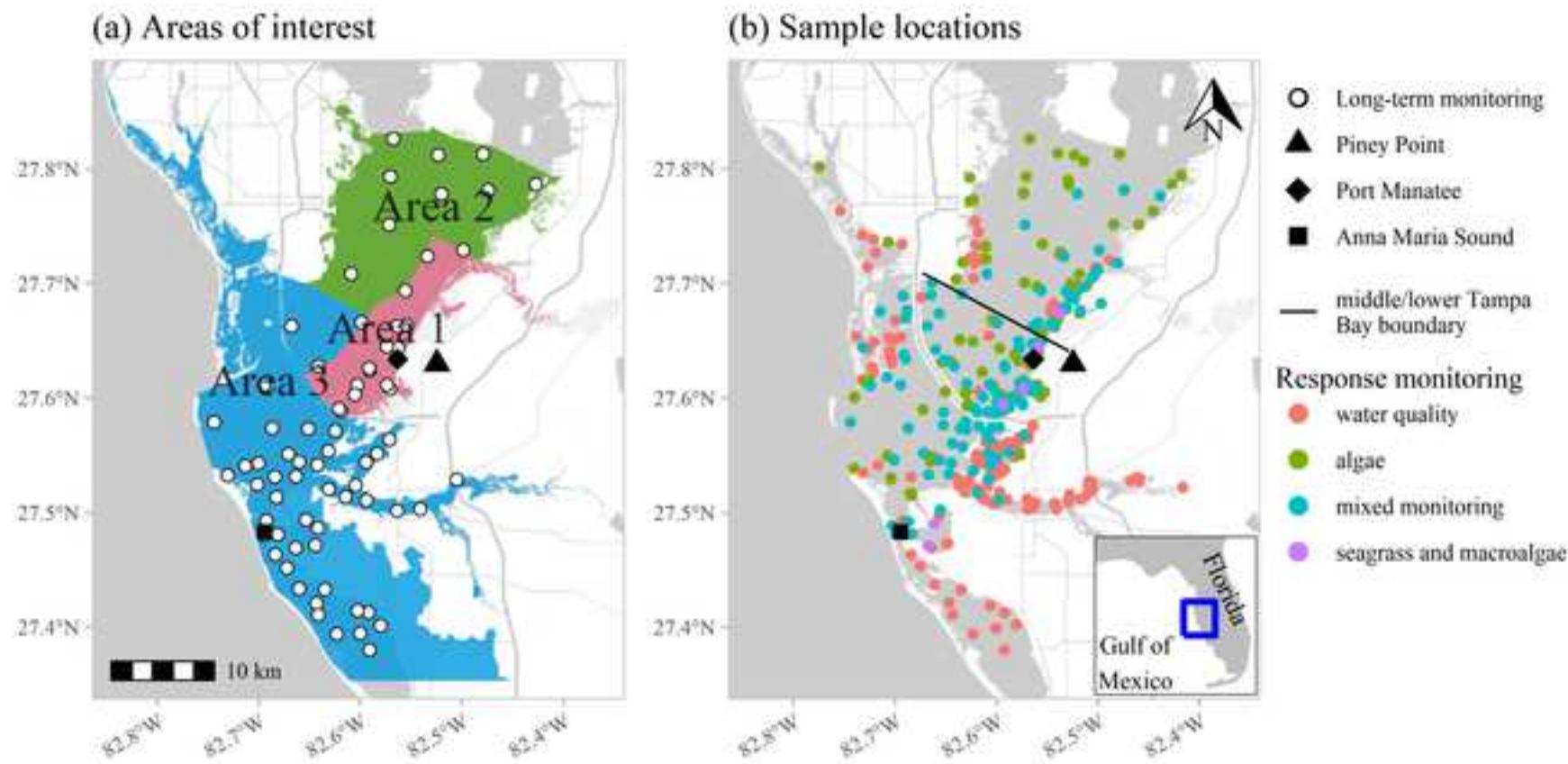
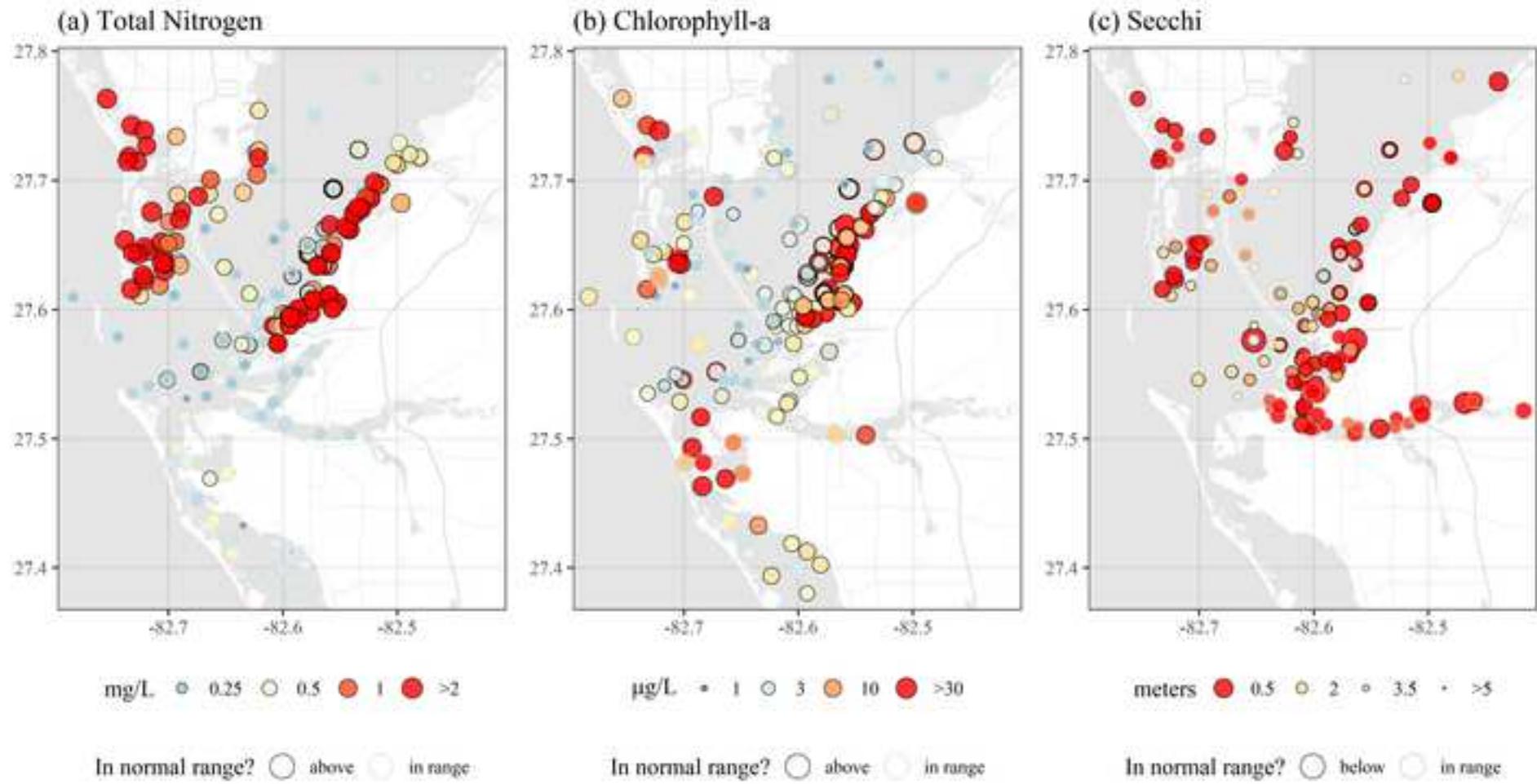
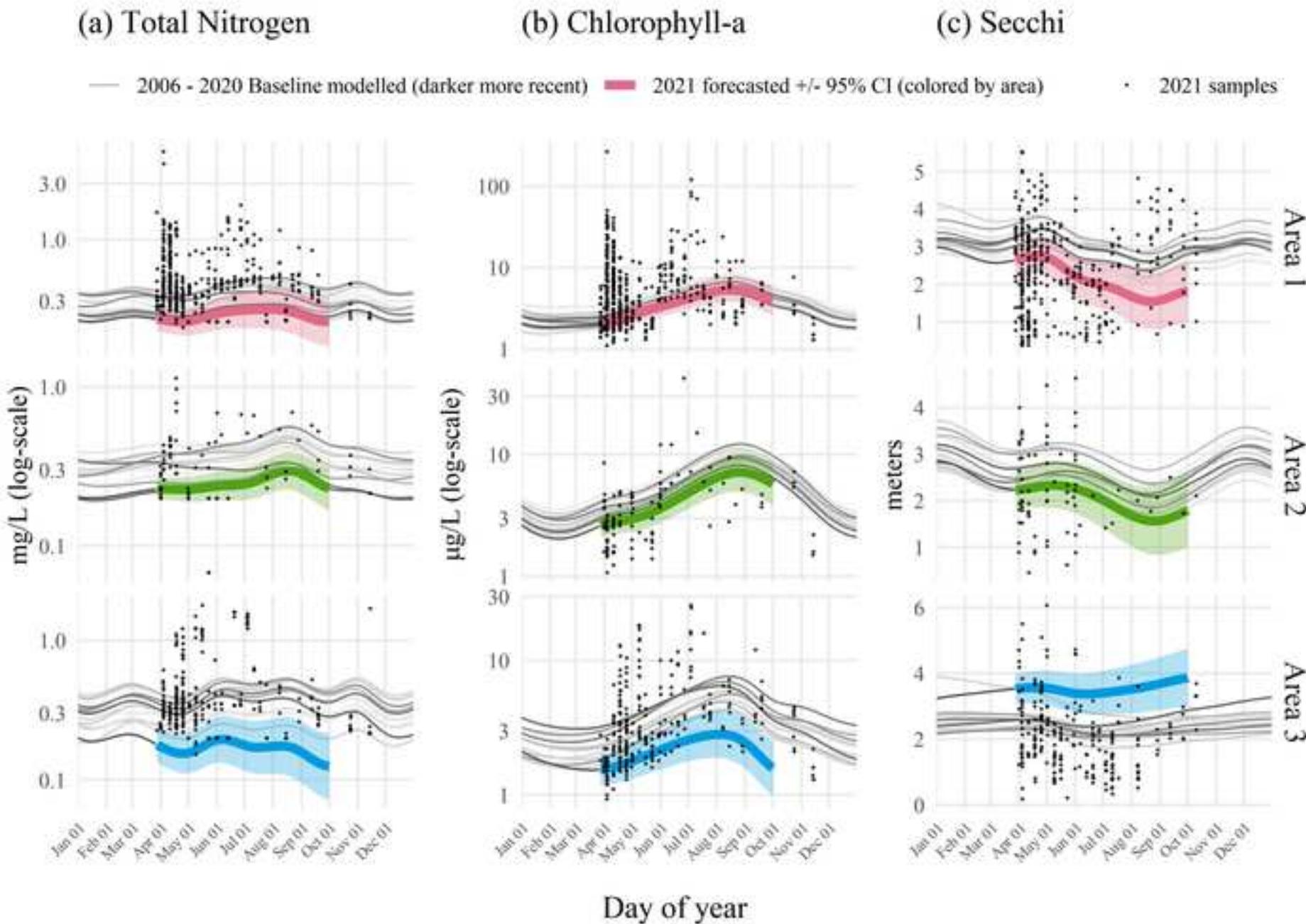
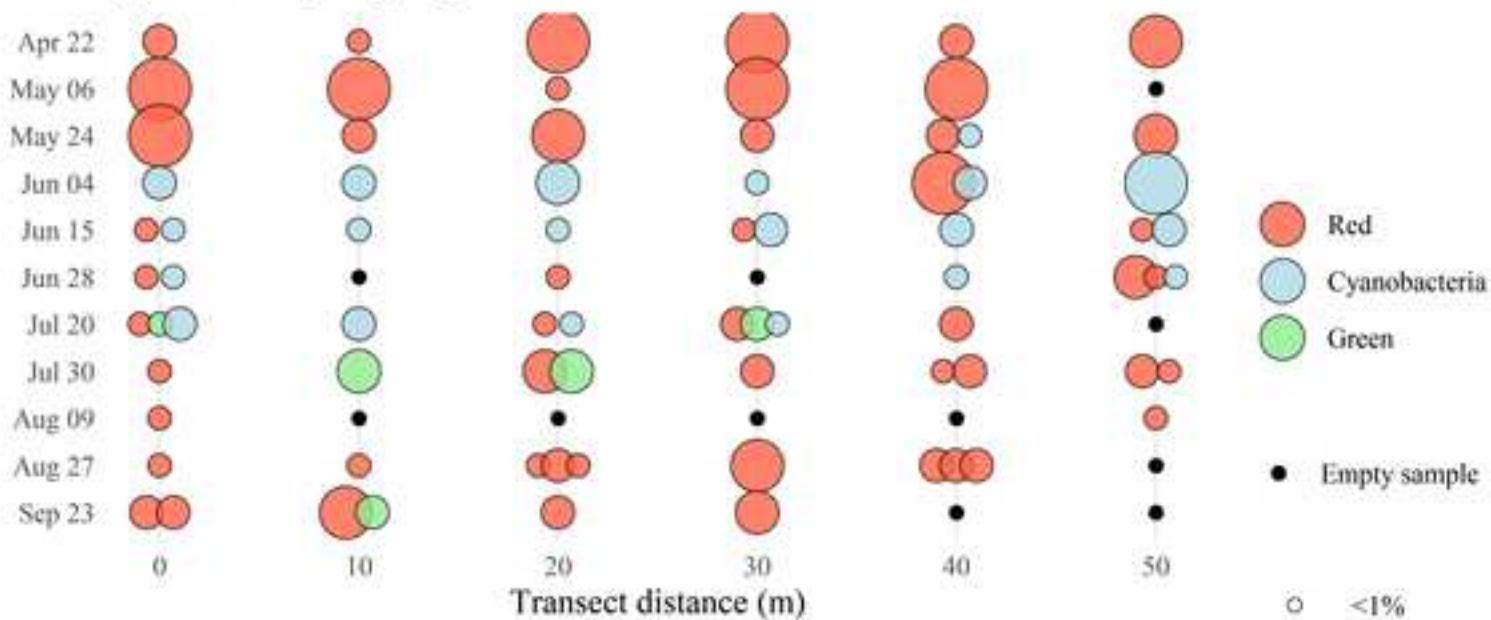


Figure 2

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## (a) Macroalgae groups



## (b) Seagrasses

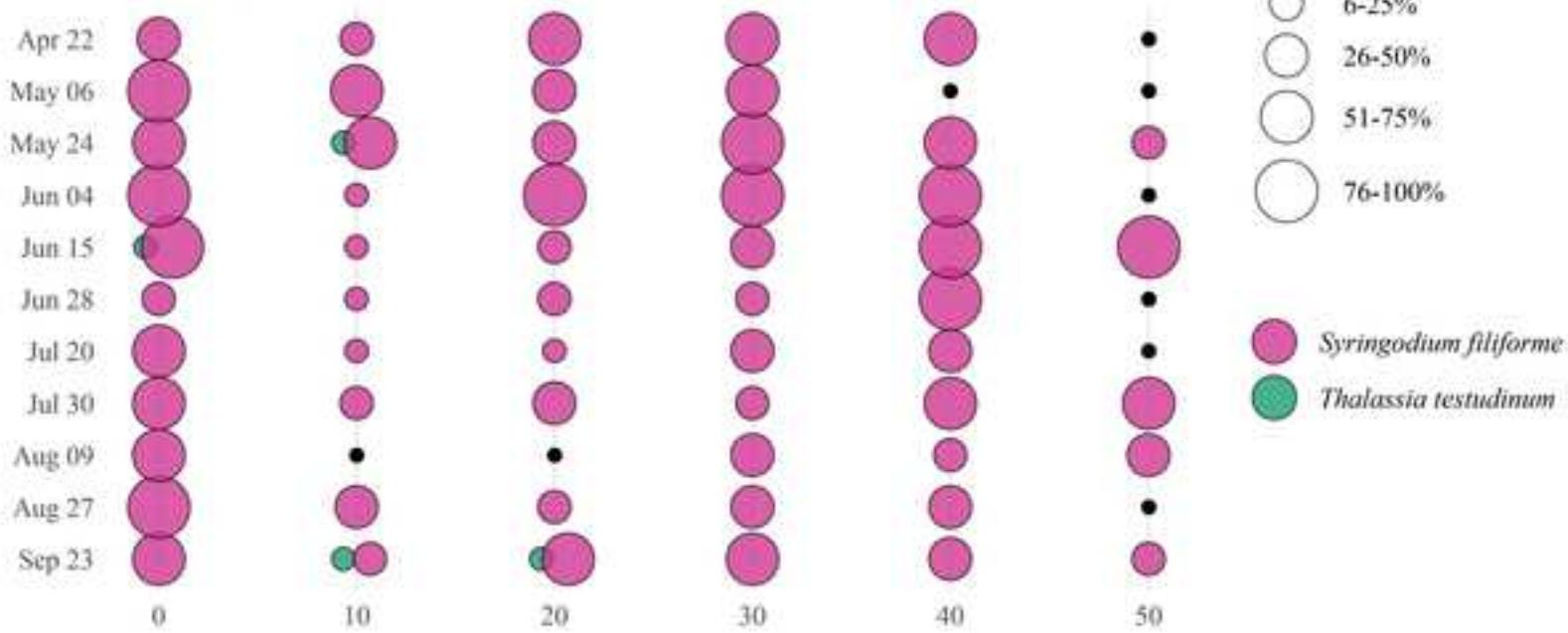
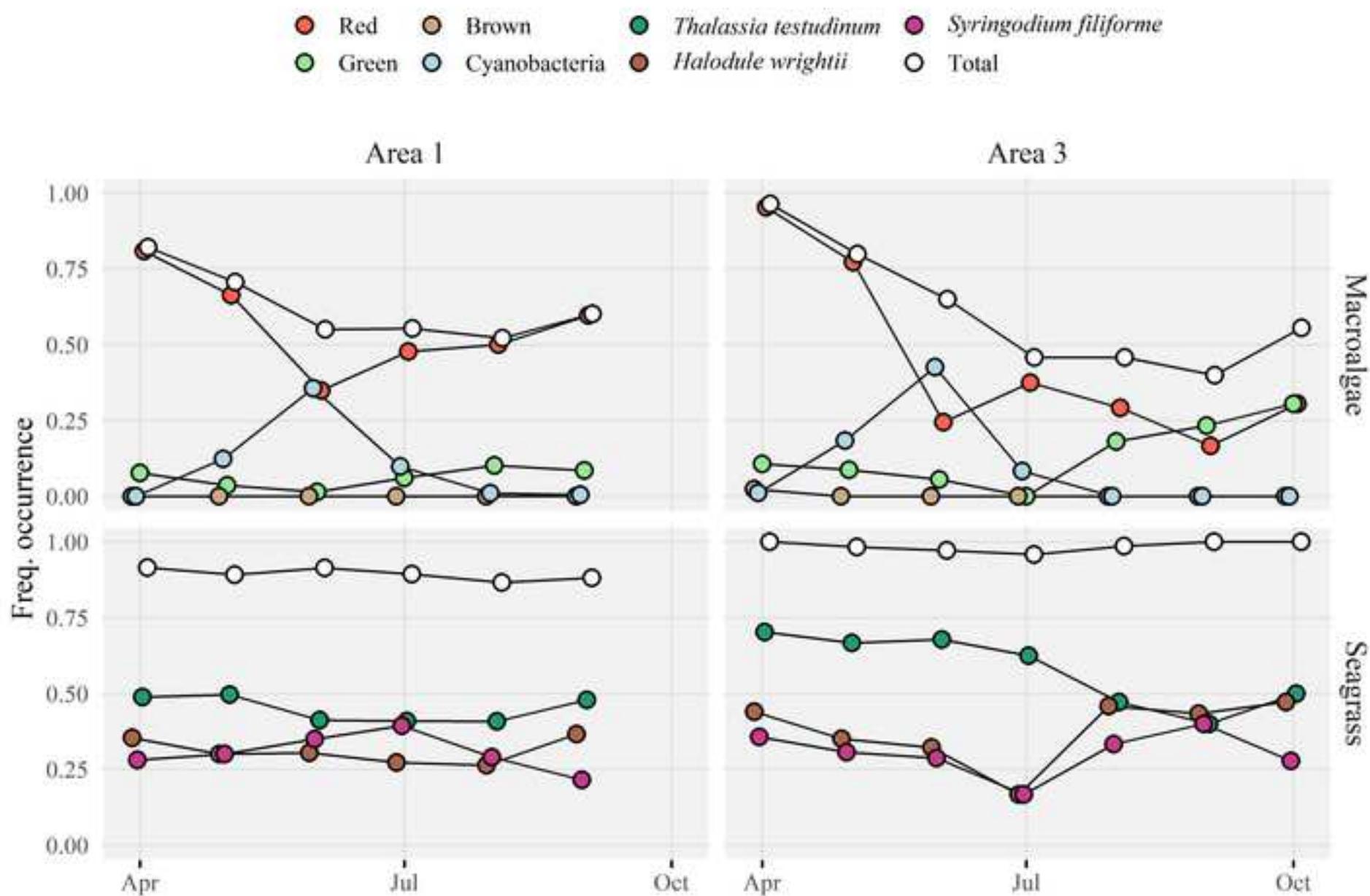


Figure 5

[Click here to access/download Figure\(s\); Fig5.jpeg](#)

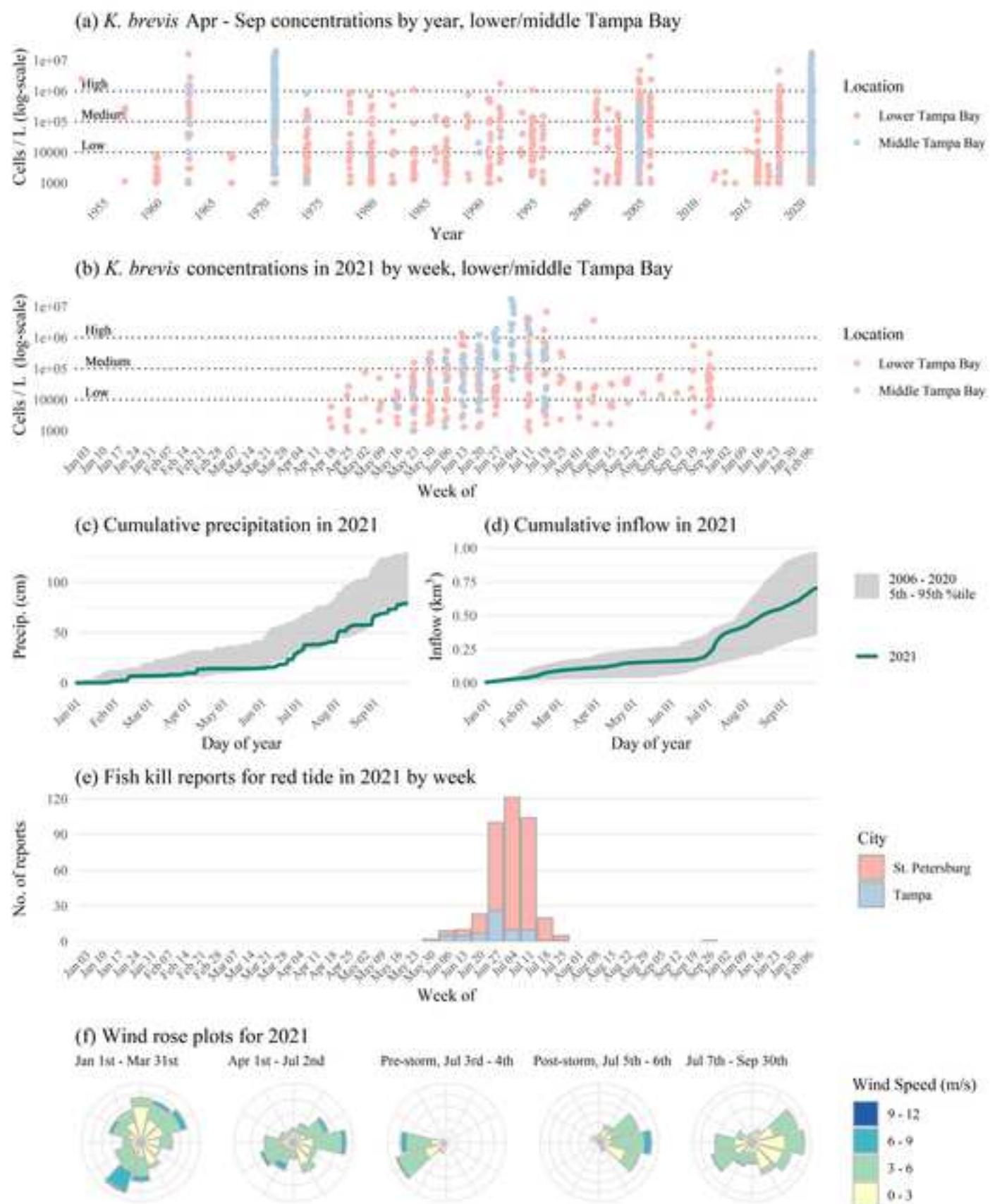
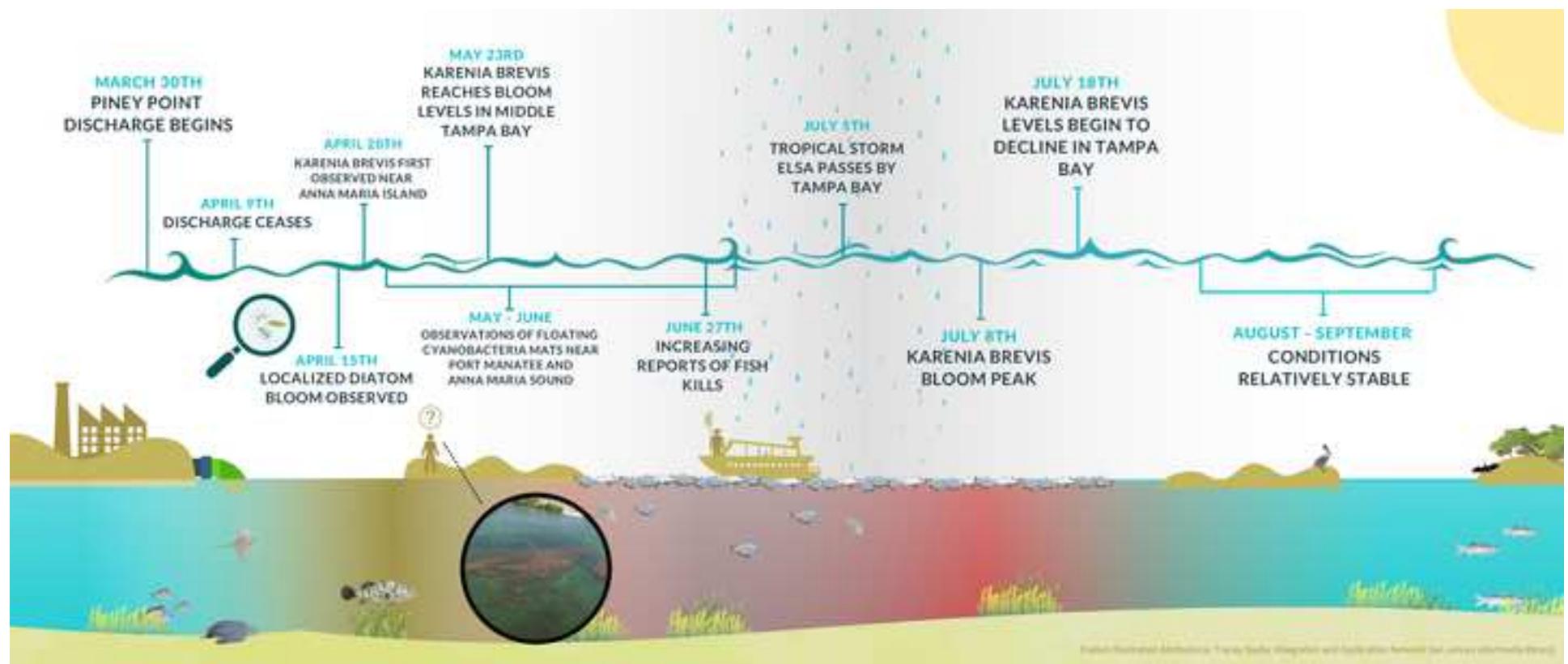
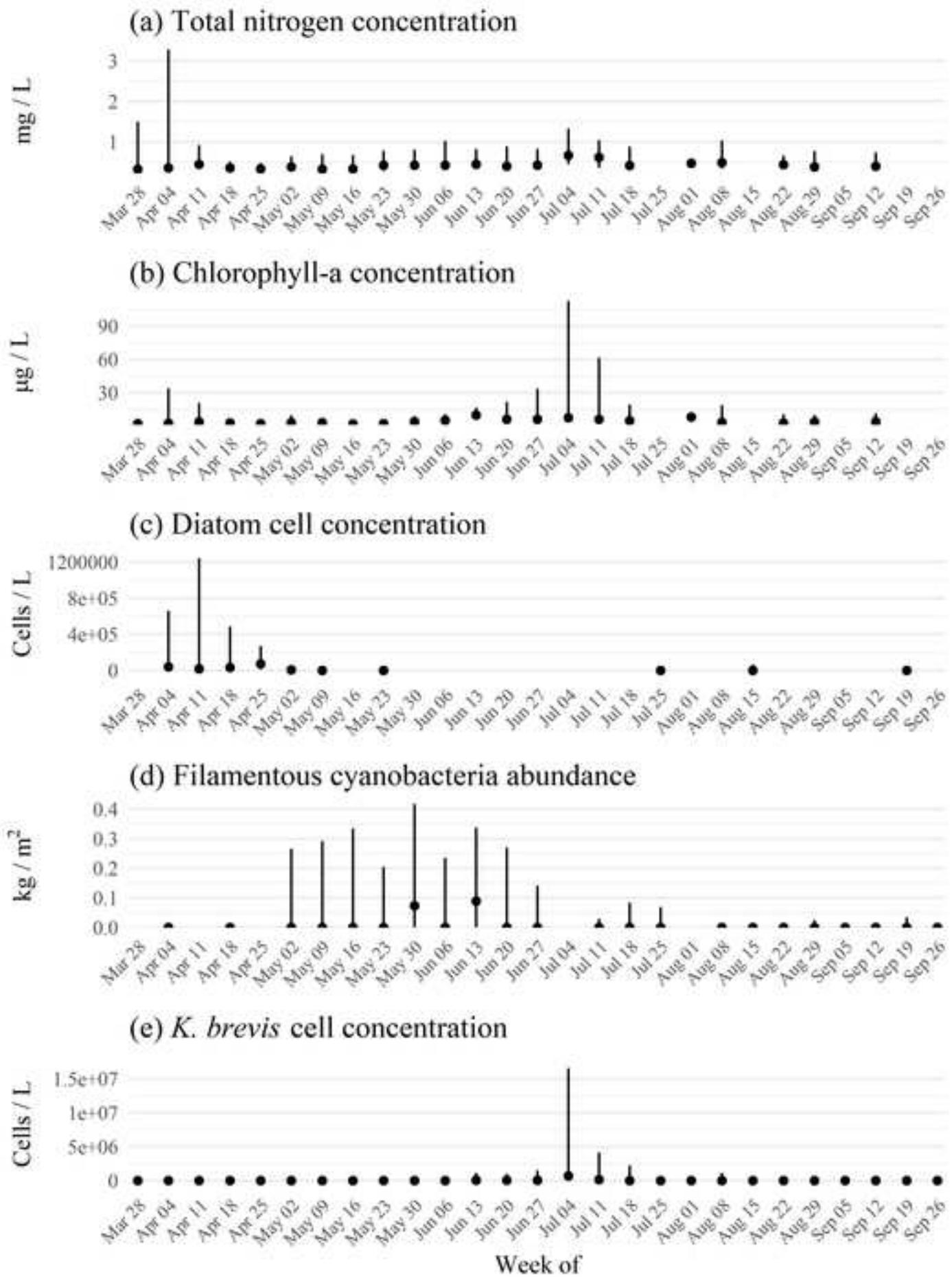


Figure 7

[Click here to access/download;Figure\(s\);Fig7.jpg](#)





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**Supplementary Data**  
supplement-revised.docx

**Declaration of interests**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

**Marcus W. Beck:** Conceptualization, Data curation, Formal analysis, Methodology, Writing – original draft, Writing – review & editing, **Andrew Altieri:** Data curation, Writing – review & editing, **Christine Angelini:** Conceptualization, Writing – review & editing, **Maya C. Burke:** Project administration, Supervision, Writing – review & editing, **Jing Chen:** Data curation, Writing – review & editing, **Diana W. Chin:** Data curation, Writing – review & editing, **Jayne Gardiner:** Data curation, **Katherine A. Hubbard:** Data curation, Validation, Writing – review & editing, **Yonggang Liu:** Data curation, Writing – review & editing, **Cary Lopez:** Data curation, Validation, Visualization, Writing – review & editing, **Miles Medina:** Formal analysis, Methodology, Visualization, Writing – review & editing, **Elise Morrison:** Date curation, Funding acquisition, Writing – review & editing, **Edward J. Phlips:** Date curation, Funding acquisition, Writing – review & editing, **Gary E. Raulerson:** Date curation, Writing – review & editing, **Sheila Scolaro:** Date curation, Writing – review & editing, **Edward T. Sherwood:** Project administration, Supervision, Writing – review & editing, **David Tomasko:** Conceptualization, Data curation, Writing – review & editing, **Robert H. Weisberg:** Date curation, Writing – review & editing, **Joe Whalen:** Conceptualization, Visualization