Understanding phytoplankton community composition in Charleston Harbor, SC using a multi-method approach

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Background

- Phytoplankton are single-celled primary producers essential to the functioning of world-wide ecosystems
- Phytoplankton communities are modified in response to abiotic conditions and serve as a proxy for environmental and anthropogenic induced change
- Estuaries are likely to be one of the first places to observe effects of climate change, such as sea level rise and increased storm flooding
- Estuaries are especially vulnerable to anthropogenically induced events such as increased storm runoff and nutrient loading (eutrophication), and harmful algal blooms (HABs).

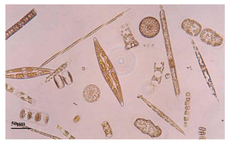

Figure 1: Light micrograph of phytoplankton.

Method Background

Microscopy
- Pros: most traditional technique, reliability
- Cons: biases large and distinct cell types, high effort, human error

High Performance Liquid Chromatography (HPLC)
- Pros: relies on pigments over direct identification, low effort
- Cons: only ID to class level, variability in pigment concentrations, issues with CHEMTAX software

Metabarcoding
- Pros: unambiguous ID to genus level, high throughput
- Cons: still in development, issues with estimation of relative abundance

A)

B)

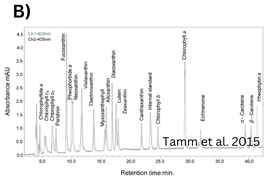

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Figure 2: A) microscope cartoon; B) example HPLC chromatogram from Tamm et al. 2015; C) illustration of DNA.

Objectives

1) Examine the phytoplankton community composition, abundance, and diversity in Charleston Harbor

2) Evaluate and examine existing methods for detecting phytoplankton communities.

Sample Collection

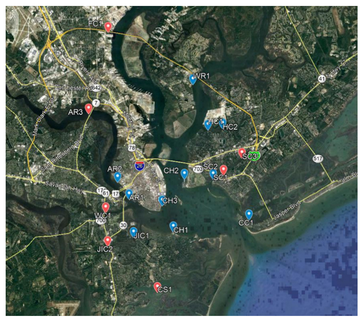


Figure 3: Map of CWK sample sites. CH1: Melton Peter Demetre Park; CH2: CofC Sailing; JIC1: James Island Creek 1; SC1; Shem Creek Park Dock.

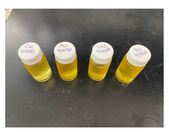
- Subset of field sites monitored by the local nonprofit Charleston Waterkeeper (CWK)
- Weekly water samples in and around Charleston Harbor, May-October 2023
- Focusing on four sites: two estuarine-based sites (CH1 & CH2), two riverine sites (JIC1& SC1)
- Only considering samples collected on high tide interval (+3ft MLLW)

Methods

- All samples are pre-filtered through 63um mesh to limit sediment contamination and interference
- **Microscopy:** fixed with 1% v/v acidic Lugol's solution (Figure 4B), identified and enumerated with an inverted microscope
- **HPLC:** HPLC and CHEMTAX preparation and analysis following Stephens et al. (*in press*)
- **Metabarcoding:** vacuum filtration onto 0.2um PVDF filters using 6-branch manifold (Figure 4A), DNA extraction using Qiagen DNeasy PowerSoil Kit, primers designed to amplify 18S rRNA and 16S rRNA genes for eukaryotes and prokaryotes respectively, sequencing using Illumina MiSeq, and taxonomic assignment using SILVA and CyanoSeq

A)

B)



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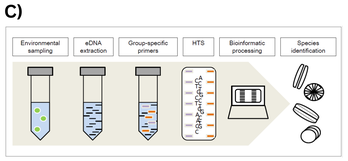

Figure 4: A) 6-branch manifold setup in laminar flow hood; B) 20mL glass vials with acidic lugol's for microscopy; C) flowchart of metabarcoding analysis.

Table 3: Quantitative metrics that can be compared across certain methods. Proxies are the outputs of each method and the dependent variables are the quantitative metric. Dependent variables are color coded to show which variables can be compared across methods. ASV = amplicon sequence variant.

	HPLC	Metabarcoding	Microscopy
Proxy	pigment ratios	ratios of DNA sequence reads	Cell counts
	pigment concentrations	DNA sequence identification	cell ID
Dependent Variable	% total by class level	% total by class level	% total by class level
		% total by ASV/genus	% total by ASV/genus
	Richness	Richness	Richness
	Alpha Diversity	Alpha Diversity	Alpha Diversity
		Beta Diversity	Beta Diversity
		% total by family	% total by family
			Cell counts total

Preliminary Results

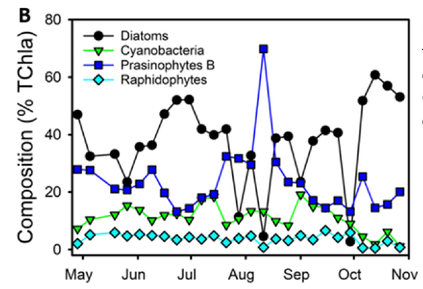

Figure 5: Community composition as the percent of TChla contributed by each algal group, as determined by CHEMTAX analysis. From Stephens et al. *in press*.

- Diatoms usually dominant, decline and replaced by Prasinophytes in late summer corresponding to increased temperature
- Cyanobacteria and raphidophytes consistent lower concentrations

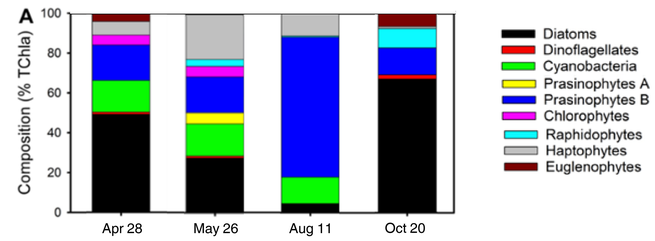

Figure 6. Stacked bar graphs of community composition as the percent of TChla contributed by each algal group from SC1. Data was taken at high tide. Data from CWK 2021. Stephens et al. *in press*.

- Increased dominance of opportunistic species in August
  - Corresponds to decreased diversity
- Demonstrates need for more regular sampling

Significance

- Essential to study response of estuaries to climate change
  - Monitoring of HABs, estuarine heat waves, eutrophication
  - Understanding synergistic stressors
- Lack of studies in developing Charleston Harbor estuary
- Evaluating methods for reliability, using multiple techniques to eliminate potential errors and provide a higher confidence in results

References & Acknowledgements

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