

# Project Notes:

## Project Title: Computational Modeling of Phytoplankton Dynamics with Climatic and Ecological Ramifications

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Note Well: There are NO SHORT-cuts to reading journal articles and taking notes from them. Comprehension is paramount. You will most likely need to read it several times, so set aside enough time in your schedule.

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## Knowledge Gaps:

This list provides a brief overview of the major knowledge gaps for this project, how they were resolved and where to find the information.

Knowledge Gap	Resolved By	Information is located	Date resolved
What oceanic variables and their impact on phytoplankton have had less research done?	Conducting review of literature	Article Notes #2, 5, 6	10 September 2023
How are ocean, climate, and phytoplankton dynamics computationally modeled?	Searching for and reading journal articles	Article Notes #7, 10	15 September 2023
How does ocean acidification impact primary production and other factors of phytoplankton?	Conducting searches for articles on Google Scholar and Scopus	Article Notes #13, 14	18 Septmeber 2023
What impacts do micronutrients have on phytoplankton populations?	Conducting searches for articles on Google Scholar and Scopus	Article #11	23 Septmber 2023
How are global warming-induced changes in oceanic conditions impacting phytoplankton at a biochemical level?	Conducting searches for articles on Google Scholar and Scopus	Article #8, 9, 14	28 September 2023
How do varying concentrations of phosphorus impact metabolic rates in phytoplankton?	Conducting searches for articles on Google Scholar and Scopus	Article #9	7 October 2023
How can computational models be validated (beyond just phytoplankton)? How is data properly put into these models?	Conducting searches for articles on Google Scholar, SCOPUS, and other databases	Article #10	14 October 2023
How can driving parameters in phytoplankton be identified? Moreover, how can interparameter relationships be established?	Conducting searches for articles on Google Scholar, SCOPUS, PubMed, ScienceDirect, and other databases	Article #10, 11, 12, 17, 18	2 November 2023
What is the relationship between rising ocean temperatures and phytoplankton dynamics	""	Article #13	15 November 2023

(particularity growth rate/abundance)?			
How can food webs be computationally modelled?	“”	Articles #15, 19	26 November 2023
What role do historical trends, and in particular, climate modelling play with regards to phytoplankton simulation?	“”	Articles #16, 20	9 December 2023

## Literature Search Parameters:

These searches were performed between 7/10/2023 and 02/01/2024.

List of keywords and databases used during this project.

Database/search engine	Keywords	Summary of search
Impact of climate change on ocean currents across multiple science news websites.	Thermohaline Circulation, Global Warming, Gulf Stream	This search was conducted across multiple science news websites including Quanta Magazine, Live Science, and phys.org. However, no satisfactory results were yielded, so searches were conducted on other news sites. Eventually, an article from Inside Climate News was chosen.
How global warming is impacting phytoplankton on Google Scholar	Phytoplankton, Global Warming, Ocean Conditions	This search revealed numerous results. The aim was to find an article offering a summative overview of what phytoplankton were, their role was in the climate system, and the impact of changing ocean conditions. One article that achieved this purpose was chosen accordingly.
Modeling carbon cycle and the role phytoplankton play in it across various science news websites.	Carbon Cycle, Biological Pump, Phytoplankton	Searches across Inside Climate News, Live Science, phys.org and Quanta Magazine provided in a variety of results. In order to understand the role of phytoplankton in the carbon cycle at a deeper level, an article from phys.org was chosen.
How do varying soil conditions brought about by different types of compost impact the concentration of nitrogen-fixing bacteria on Google Scholar	Nitrogen-Fixing Bacteria, Soil Conditions, Composting	A myriad of results were provided from the search, all relating to some facet of composting and/or nitrogen-fixing bacteria. One article regarding the concentration of antibiotics in composting was of interest, and so it was read.
Another, more deeper search on Google Scholar about the impact of climate	Ocean Conditions, Climate Change, Phytoplankton, Primary	The aim with this search was to focus more specifically on the changes phytoplankton are

change on phytoplankton	Production, Light, Temperature	facing, what adaptations they may be performing, and how this may vary region to region. From this, two articles were chosen: one relating to the adaptations and changes phytoplankton are undergoing, and one that focuses on the impact of changing conditions in the Southern Ocean specifically.
Modeling the impact of changing ocean conditions on Scopus	Ocean Modeling, Phytoplankton, Climate Change	One area of consideration for the STEM research project was to develop a computer model to track how phytoplankton will be impacted by ocean conditions. Conducting this search on Scopus yielded various articles, including one about modeling changes for zooplankton. This article was chosen to be read in order to gain better insight into how modelling phytoplankton may end up working. Another article that discussed phytoplankton directly was also chosen for further investigation.
Ecological services offered by phytoplankton on Scopus	Ecological Services, Phytoplankton	This search was conducted with the purpose of attaining an article that could easily be cited when discussing the importance of phytoplankton during work on the STEM project. This search resulted in various articles, and the one that appeared to most robustly delineate the ecological services of phytoplankton was chosen.
How is global warming changing the biochemistry of phytoplankton on Scopus	Biochemistry, Metabolism, Primary Production, Phytoplankton, Global Warming	The biochemistry involved in the impact of changing oceans on phytoplankton was another angle of consideration for the STEM project. This search provided a wide variety of results, varying from chlorophyll concentrations, genetic conditions, and other factors. From this article regarding the potential positive impact of DNA methylation and gene transcription was chosen to be read.
How is ocean acidification impacting the metabolism of phytoplankton on Scopus	Ocean Acidification, Metabolism, Phytoplankton	This search was aimed at investigating the relationship between a specific oceanic condition and some facet of phytoplankton. For this search, the impact of ocean acidification on metabolism was investigated, and one article that offered a good overview of this relationship was chosen to be read.
What impacts do varying levels of micronutrients have on phytoplankton? On Google Scholar, SCOPUS, PubMed,	Micronutrients, Phytoplankton, Biochemistry,	This search was conducted with the ends of developing a deeper understanding of the relationship between micronutrients and

ScienceDirect, and other databases (hereafter: on “”)		phytoplankton populations. A study with comprehensive coverage of micronutrients, as well as DO, chlorophyll, temperature, and other variables in Bangladeshi coastal waters was selected to be read.
How do varying levels of phosphorus impact metabolic rates in phytoplankton? On “”	Phytoplankton, metabolism, phosphorus	This search was aimed at investigating the relationship phosphorus holds with metabolic rates in phytoplankton populations. It was hypothesized that greater phosphorous levels would increase metabolism. To evaluate this hypothesis, the literature was reviewed, and a very recent article discussing how varying levels of phytic acid impact metabolic rates was discussed.
A concrete example of computational modelling a real-life system On “”	Phytoplankton, computer modelling	This search was conducted in order to gain a more developed background in computational modeling techniques, specifically with regards to phytoplankton. Two articles, one more general, the other focusing in on a particular reservoir in Brazil, were chosen to be read.
AI modelling techniques and phytoplankton On “”	KNNs, Network theory, louvonian method,	A wide array of models for varying purposes were found from this search, ranging from food webs to time series, driving parameter and climatic models, among other tools. These studies were chosen to be analyzed.
Computational Food Webs On “”	Network theory, phytoplankton dynamics, food webds	This search offered numerous case-studies of ecosystems whose food webs were modelled. Said studies were analyzed.
Climare models on On “”	CMIP5, phytoplankton	This search produced some results regarding the specified and model and phytoplankton. Relevant articles were chosen to be read.

## Tags:

Tag Name	
<a href="#">Description of and Example of Change in Thermohaline Cycling</a>	<a href="#">Example of the Importance of Accurate and Precise Modeling and the Drastic Impact of Minor Changes to Model Parameters</a>
<a href="#">Possible Laboratory Techniques to Perform on Phytoplankton</a>	<a href="#">Definitions of Major Processes in the Nitrogen Cycle</a>

<a href="#"><u>qPCR (Quantitative Polymerase Chain Reaction) Defintion</u></a>	<a href="#"><u>Description of DGGE (Denaturing Gradient Gel Electrophoresis)</u></a>
<a href="#"><u>Impact of Climate Change on Ocean Conditions</u></a>	<a href="#"><u>Shifting Competitvie Advantages for Phytoplankton</u></a>
<a href="#"><u>Use of Time Series as a Tool for Oceanic Modeling</u></a>	<a href="#"><u>Comparing Results of Different Computer Modeling Techniques and Identifying Methods that Optimize Prediction Accuracy</u></a>
<a href="#"><u>Various Parameters In Modeling Phytoplankton Populations</u></a>	<a href="#"><u>Considering Degree of Gelatiny as a Possible Area of Study</u></a>
<a href="#"><u>Possible Tool of DNA Metabarcoding</u></a>	<a href="#"><u>Transferrance of Fatty Acids Up the Trophic Pyramid (focusing on cycling and dynamics of specific substances in phytoplankton)</u></a>
<a href="#"><u>Role of Phytoplankton in Biogenic Sulfur Budget Explanation</u></a>	<a href="#"><u>Metabolism of Various Macromolecules</u></a>
<a href="#"><u>Fundamental Gene Expression Analysis Process</u></a>	<a href="#"><u>Investigating the Impact of Heavy Metals and Proteins on Phytoplankton and Drawing Connections to Phosphates and Other Compounds</u></a>
<a href="#"><u>Metabolic Ramifications for Higher-Order Organisms</u></a>	<a href="#"><u>Comparing Results of Different Computer Modeling Techniques and Identifying</u></a>
<a href="#"><u>Methods that Optimize Prediction Accuracy</u></a>	<a href="#"><u>Various Parameters In Modeling Phytoplankton Populations</u></a>
<a href="#"><u>Advanced Statistical Tests and Techniques</u></a>	<a href="#"><u>Drivers of Phytoplankton Dynamics Globally</u></a>
<a href="#"><u>Potential Source of Raw, Actual Data on Environmental Factoes Influencing Phytoplankton Dynamics</u></a>	<a href="#"><u>Searching for Factors Impacting Carbon Sequestration</u></a>
<a href="#"><u>Possible Biotechnological Connections</u></a>	<a href="#"><u>Definition of Four Major Groups of Phytoplankton</u></a>
<a href="#"><u>Alternative General Data Analysis Software to Sheets or Excel</u></a>	<a href="#"><u>Basis for Thermal Equation and Criteria Used in Study</u></a>
<a href="#"><u>Basic Outline of Fatty Acid Metabolism</u></a>	<a href="#"><u>Key Attributes of a Food Web</u></a>

<a href="#">Basic Computational Construction of a Food Web</a>	<a href="#">Relevant Instances of Computational Limitations</a>
<a href="#">Potential Data Analysis Tool (Analogous to December Fair)</a>	<a href="#">Means of Measuring Ecological Diversity</a>
<a href="#">Another, More Detailed Example of Computational Food Web Construction</a>	<a href="#">An Example Of Phytoplankton-Climate Relations</a>
<a href="#">Instance of using chlorophyll as an indicator for phytoplankton dynamics</a>	

## Template for Article Notes

Article notes should be on separate sheets

**KEEP THIS BLANK AND USE AS A TEMPLATE**

<b>Source Title</b>	
<b>Source citation (APA Format)</b>	
<b>Original URL</b>	
<b>Source type</b>	
<b>Keywords</b>	
<b>#Tags</b>	
<b>Summary of key points + notes (include methodology)</b>	
<b>Research Question/Problem/Need</b>	
<b>Important Figures</b>	
<b>VOCAB: (w/definition)</b>	
<b>Cited references to follow up on</b>	
<b>Follow up Questions</b>	

NOTES

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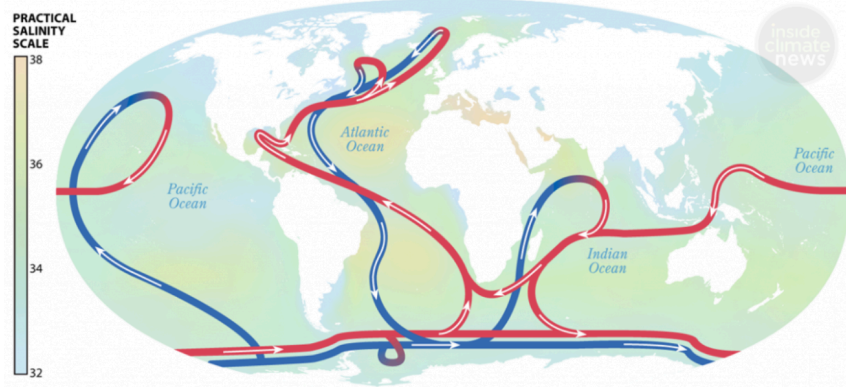
## Article #1 Notes: Scientists Say Ocean Circulation Is Slowing. Here's Why You Should Care.

<b>Source Title</b>	Scientists Say Ocean Circulation Is Slowing. Here's Why You Should Care.
<b>Source citation (APA Format)</b>	Berwyn, B. (2018, May 7). <i>Scientists Say Ocean Circulation Is Slowing. Here's Why You Should Care</i> . Inside Climate News. <a href="https://insideclimatenews.org/news/07052018/atlantic-ocean-circulation-slowing-climate-change-heat-temperature-rainfall-fish-why-you-should-care/?gclid=CjwKCAjw2K6lBhBXEiwA5RjtCZMQVm0nxKHzSuutQ_Cgz9mZ1peI8xnzAhVnN0VCr8vudXHq2Sa3IhoC5O0QAvD_BwE">https://insideclimatenews.org/news/07052018/atlantic-ocean-circulation-slowing-climate-change-heat-temperature-rainfall-fish-why-you-should-care/?gclid=CjwKCAjw2K6lBhBXEiwA5RjtCZMQVm0nxKHzSuutQ_Cgz9mZ1peI8xnzAhVnN0VCr8vudXHq2Sa3IhoC5O0QAvD_BwE</a>
<b>Original URL</b>	<a href="https://insideclimatenews.org/news/07052018/atlantic-ocean-circulation-slowing-climate-change-heat-temperature-rainfall-fish-why-you-should-care/?gclid=CjwKCAjw2K6lBhBXEiwA5RjtCZMQVm0nxKHzSuutQ_Cgz9mZ1peI8xnzAhVnN0VCr8vudXHq2Sa3IhoC5O0QAvD_BwE">https://insideclimatenews.org/news/07052018/atlantic-ocean-circulation-slowing-climate-change-heat-temperature-rainfall-fish-why-you-should-care/?gclid=CjwKCAjw2K6lBhBXEiwA5RjtCZMQVm0nxKHzSuutQ_Cgz9mZ1peI8xnzAhVnN0VCr8vudXHq2Sa3IhoC5O0QAvD_BwE</a>
<b>Source type</b>	Scientific News Article
<b>Keywords</b>	No keywords are given.
<b>#Tags</b>	<a href="#">Description of and Example of Change in Thermohaline Cycling</a>
<b>Summary of key points + notes (include methodology)</b>	The rate at which the thermohaline cycle operates, that is, the rate at which warmer, fresher water evaporates and as a result cools and sinks as it becomes more saline and dense, is slowing due to warmer ocean temperatures, leading to slower ocean currents. Although the ramifications of this are not fully known, based off data about past climate conditions, these trends in ocean currents have been associated with extreme climate patterns. The article emphasizes the need for further investigation into the impacts of changing ocean currents.
<b>Research Question/Problem/Need</b>	How is climate change impacting ocean currents, and what climatic and/or environmental implications could this have?

## Important Figures

**Thermohaline Circulation: The Great Ocean Conveyor Belt**

Currents circulate water through the world's oceans like a giant conveyor belt, carrying heat from the tropics toward the poles and sending colder water back in deep ocean currents. Wind, temperature and salinity help drive them. This simplified map shows the pattern.



This figure depicts the thermohaline cycle in action, as well as ocean salinity. Arrows are used to indicate the direction of the current. Red lines represent the warm, fresh, less dense water at the surface of the ocean. Blue lines represent deeper, cooler, denser, and saltier ocean waters. Analysis of the color of the arrows throughout the globe reveals that the northeast Atlantic and western Antarctic are major sites for where water sinks, whereas the northwestern Indian Ocean and Pacific are major sites for ocean water rising. Meanwhile, salinity is measured using the practical salinity scale, the ratio between the conductivity of a given seawater sample and that of a standard solution, where the standard solution has a salinity of about 35. Bluer shade indicate lower salinity, while the more orange ones indicate higher ones. Ocean waters at the poles and equator appear to be less saline, while salinity is higher for ocean waters at the intermediate latitudes. The bodies of water with the highest salinity seem to be the Mediterranean and Red Seas, as well as the Persian Gulf.

**VOCAB: (w/definition)**

**Thermohaline Cycling-** A major process by which ocean currents operate. Water at the surface of the ocean is warmer and more fresh, meaning it has fewer salts dissolved in it, and therefore less dense. However, this warm water evaporates, leaving behind ocean salts in the remaining water. The water therefore becomes cooler, denser, and more saline, and sinks. However, this water eventually returns to the surface as heat from the Earth's core increases its temperatures, and thus decreases its salinity and density.

**Practical Salinity Scale-** Means by which salinity in ocean water is measured. Quantitatively, this is the ratio between the conductivity of the seawater sample and the conductivity of a standard solution that contains 1 kg of solution, and about 32.4 g of Potassium Chloride at 15 °C.

**Cited references to follow up on**

Caesar, L., Rahmstorf, S., Robinson, A., Feulner, G., & Saba, V. (2018). Observed fingerprint of a weakening Atlantic Ocean Overturning Circulation. *Nature*, 556(7700), 191–196.  
<https://doi.org/10.1038/s41586-018-0006-5>

	<p>Valley, S., Lynch-Stieglitz, J., &amp; Marchitto, T. M. (2017). Timing of deglacial AMOC variability from a high-resolution seawater cadmium reconstruction. <i>Paleoceanography</i>, 32(11), 1195–1203.  <a href="https://doi.org/10.1002/2017pa003099">https://doi.org/10.1002/2017pa003099</a></p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How do oceanic pH conditions impact the speed of the thermohaline cycle? What about the concentrations of certain nutrients and other chemicals? How do these different variables interact in tandem to impact ocean currents?</li> <li>2. How do slower ocean currents impact the severity of storms, and how does that impact ocean conditions in turn? Is there a possible feedback loop between ocean currents, weather conditions, and ocean conditions?</li> <li>3. How will slower ocean currents impact interactions across the food web?</li> <li>4. What impact would a slower thermohaline cycle have on primary production in phytoplankton?</li> </ol>

## NOTES:

- A prospective area of focus for the STEM research project that I have relates to climatology, specifically, how global warming is impacting various elements of the global climate system, and in turn, how that may impact the climate in the future.
- This scientific news article discusses how rising temperatures are slowing the rate of ocean currents and the impacts of that.
- Thermohaline circulation plays an important role in influencing climate and transferring heat and nutrients.
- Ocean currents have slowed to unprecedented rates, while ocean temperatures, due to carbon dioxide emissions, have reached historically high levels.
- Article places a heavy focus on the Gulf Stream, which flows Northeast along the North Atlantic from the Caribbean to Europe.
- Melting from the Greenland Ice Sheet due to rising temperatures has created more freshwater at a point in the Gulf Stream where warmer and less dense water usually begins to sink as freshwater evaporates and the water becomes more saline and thus more dense, sinking and cooling. With more freshwater present, the density of the warmer surface water is decreased, slowing the process of sinking and cooling, and in turn, the rate of the current.
- The exact consequences of warmer ocean temperatures and slower currents remains unclear, although in the past, they have been associated with extreme climate patterns.
  - For example, rapidly warming water off the New England coast has negatively impacted the cod industry and led to extreme weather such as Hurricanes Irene and Sandy.
- Similar trends of warming oceans and slowing currents have been observed in other regions of the ocean across the globe. Since many of these currents are much larger, the impact on the climate may be even more drastic.

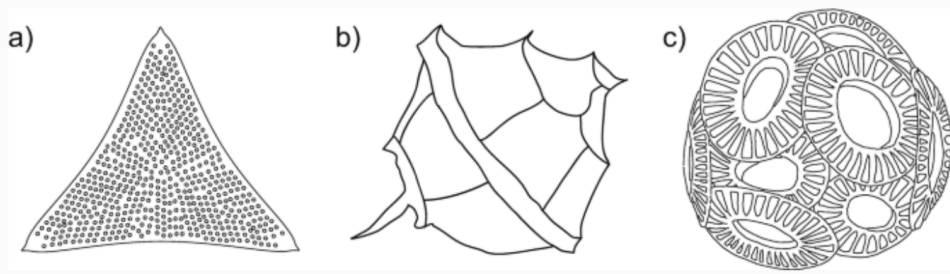
- There is a need for further research into the effects of changing ocean conditions in order to paint a clearer picture of this situation and allow communities to make more informed choices with regards to preparation and mitigation efforts.

## Article #2 Notes: Phytoplankton Responses to Marine Climate Change – An Introduction

<b>Source Title</b>	Phytoplankton Responses to Marine Climate Change – An Introduction
<b>Source citation (APA Format)</b>	Käse, L., & Geuer, J. K. (2018). Phytoplankton Responses to Marine Climate Change – An Introduction. <i>YOUMARES 8 – Oceans Across Boundaries: Learning from Each Other</i> , 55–71. <a href="https://link.springer.com/chapter/10.1007/978-3-319-93284-2_5">https://link.springer.com/chapter/10.1007/978-3-319-93284-2_5</a>
<b>Original URL</b>	<a href="https://link.springer.com/chapter/10.1007/978-3-319-93284-2_5">https://link.springer.com/chapter/10.1007/978-3-319-93284-2_5</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Phytoplankton Community, Seasonality, Harmful Algal Blooms, Time Series, Primary Production
<b>#Tags</b>	<a href="#">Possible Laboratory Techniques to Perform on Phytoplankton</a> (see figure 3 below also) <a href="#">Impact of Climate Change on Ocean Conditions</a>
<b>Summary of key points + notes (include methodology)</b>	Phytoplankton are a diverse group of microscopic autotrophs that lie at the forefront of climate regulation and at the base of marine food chains. The article discusses various laboratory techniques that can be used to analyze changes and characteristics in phytoplankton populations, and how that may be utilized to study how climate change is impacting them. There is a significant dearth of understanding regarding the impact of changing ocean conditions on phytoplankton, impacts which seem to vary drastically from region to region, meanwhile the importance of building on existing models and methods to paint a clearer picture of these dynamics is stressed.
<b>Research Question/Problem/Need</b>	Broadly speaking, how are global warming-induced changes in ocean conditions impacting phytoplankton populations, how can this be modelled and measured, and what ramifications does this have for the climate and environment?

## Important Figures

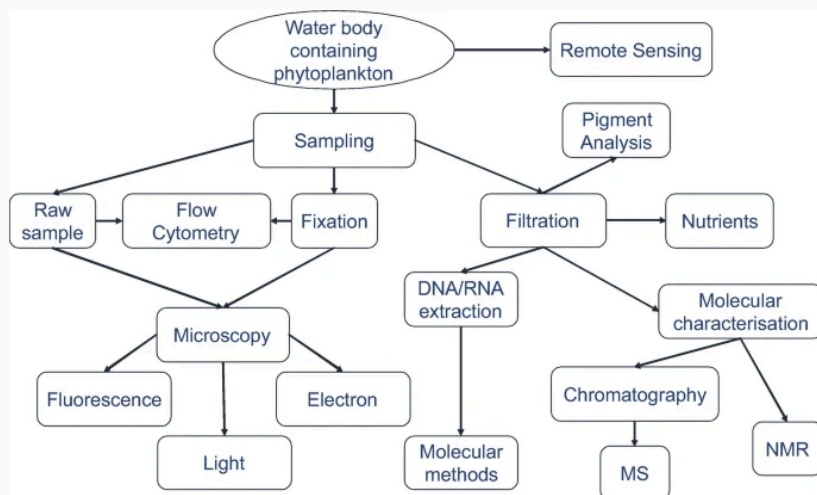
Fig. 1



Exemplary schematic drawings of three important phytoplankton groups. (a) Triangular diatom *Trigonium* sp., (b) dinoflagellate *Pyrodinium bahamense* and (c) coccolithophorid *Emiliana huxleyi* (prymnesiophytes). (Adapted from the open source Plankton\*Net Data Provider at the Alfred Wegener Insitute for Polar and Marine Research (a) and (c), and from Landsberg et al. (2006) (b))

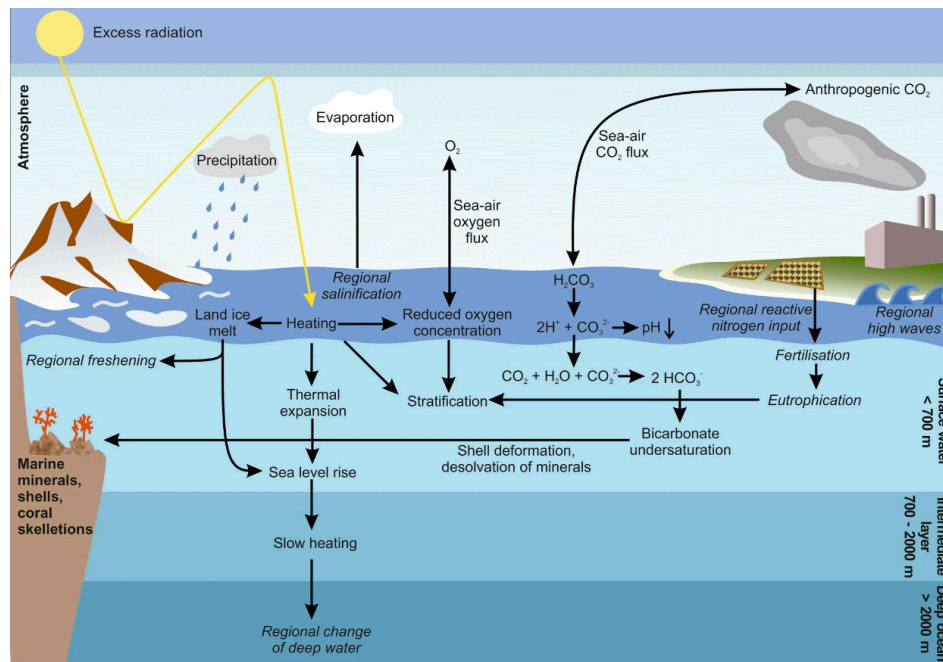
This figure illustrates the basic structure of three main groups of phytoplankton: diatoms, dinoflagellates, and coccolithophorids. All three groups vary in shape and size. The diatom is porous and triangular. The dinoflagellates, as per its name, appears to have a flagella. The coccolithophorid is also very porous, but unlike the diatom, it is comprised of many donut-like structures. The pores are located on the edges of these structures. Different shapes and structures across different groups allows for specialization in function, habitat, range of habitable conditions, and other important environmental metrics. The difference in structure among these three groups alone is an indication of the diversity present within the phytoplankton community.

Fig. 3



Schematic overview of the methods used for phytoplankton studies. Three different possibilities to process the sample are using raw samples, fixation or preservation, and filtration. For microscopy and flow cytometry raw samples either are measured immediately or have to be fixed for later measurements. Since molecular methods, pigment analysis and detection of molecular tracers usually require concentrated cells, filter residues serve for phytoplankton measurements. Molecular characterization and quantification of trace molecules is performed using chromatography, mass spectrometry (MS), and nuclear magnetic resonance (NMR) spectroscopy

This figure is a flowchart of possible methods that can be used to study phytoplankton. Given that phytoplankton are microscopic in size, most methods performed are at a microscopic scale or smaller. Even so, methods vary from chromatography and pigment analysis to mass spectrometry, methods with electrons and light, and nucleotide analyses.



Overview about climatic changes and their effects on the ocean after Ciais et al. (2013) and Rhein et al. (2013). Regional effects are displayed in italics. Excess solar radiation enters the atmosphere. Ice reflects this radiation, but it is taken up by the surface ocean, leading to its warming. Ocean warming results in land ice melt and thermal expansion, which both result in a sea level rise. Heating of vast areas of the surface ocean also slowly heats up the intermediate water layer which, among others, can ultimately lead to regional changes of deep water. Regional freshening occurs on sites with melting land ice. Regional salinification on the contrary happens in areas of vast evaporation. Surface ocean warming also decreases the solubility of gases, leading to a reduced oxygen concentration and thus changes in the sea-oxygen flux. Excess anthropogenic carbon dioxide enhances its uptake by the ocean and leads to a gradual acidification of the ocean. A decreasing pH results in bicarbonate undersaturation, which causes dissolving of shells and other minerals. Regional input of reactive nitrogen can lead to fertilization and eutrophication. Another regional effect is the occurrence of high waves. Heating, reduced oxygen concentrations and eutrophication lead to higher stratification of water masses

This figure illustrates the various anthropogenic impacts on oceanic conditions. Higher sea levels, reduced nutrient concentrations, acidification, and other changes in ocean conditions, in addition to exerting their exclusive impacts, seem to result in further stratification in the ocean. Labels that are in italics indicate regional oceanic changes. For example, due to climate change, there is an increasing contrast between fresh and saline areas, as noted above. Visualizing these changes aids in understanding them, and in turn, what that may mean for phytoplankton populations.

**VOCAB:**  
(w/definition)

**Primary Production** - The creation of energy for living organisms in the form of organic molecules. The main source of primary production on Earth is photosynthesis.

**Spectrometry** - The means by which varying wavelengths of light (commonly spanning from UV to InfraRed light) is measured. There are varying types of spectrometry that focus on lights with different frequencies, electrical charge, and other elements of quantum mechanics.

**Lysis** - In the context of biochemistry, the breaking apart of a larger molecule into smaller components.

	<b>Anoxic</b> - The absence of dissolved oxygen in the ocean.
<b>Cited references to follow up on</b>	<p>Boyd PW, Jickells T, Law CS et al (2007) Mesoscale iron enrichment experiments 1993–2005: synthesis and future directions. <i>Science</i> 315:612–617. <a href="https://doi.org/10.1126/science.1131669">https://doi.org/10.1126/science.1131669</a></p> <p>Bradley PB, Sanderson MP, Frischer ME et al (2010) Inorganic and organic nitrogen uptake by phytoplankton and heterotrophic bacteria in the stratified Mid-Atlantic Bight. <i>Estuar Coast Shelf Sci</i> 88:429–441. <a href="https://doi.org/10.1016/j.ecss.2010.02.00">https://doi.org/10.1016/j.ecss.2010.02.00</a></p> <p>Buesseler KO, Andrews JE, Pike SM et al (2004) The effects of iron fertilization. <i>Science</i> 304:414–417. <a href="https://doi.org/10.1126/science.1086895">https://doi.org/10.1126/science.1086895</a></p> <p>Etheridge SM, Roesler CS (2005) Effects of temperature, irradiance, and salinity on photosynthesis, growth rates, total toxicity, and toxin composition for <i>Alexandrium fundyense</i> isolates from the Gulf of Maine and Bay of Fundy. <i>Deep Res Pt II</i> 52:2491–2500. <a href="https://doi.org/10.1016/j.dsr2.2005.06.026">https://doi.org/10.1016/j.dsr2.2005.06.026</a></p> <p>Fenchel T (1988) Marine plankton food chains. <i>Annu Rev Ecol Syst</i> 19:19–38. <a href="https://doi.org/10.1146/annurev.es.19.110188.000315">https://doi.org/10.1146/annurev.es.19.110188.000315</a></p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How does increased oceanic stratification impact the concentrations of chlorophyll a in phytoplankton populations at different depths?</li> <li>2. How can molecular level techniques used to study phytoplankton be paired with larger scale models used for depicting ocean, atmosphere and climate conditions?</li> <li>3. Which species of phytoplankton dominate saline environments, and which ones dominate fresher ones, and how will those dynamics play out as oceans become either excessively saline or fresh?</li> <li>4. How does the shape and overall structure of phytoplankton cells impact its adaptations to global warming-induced changes in oceanic conditions?</li> </ol>

## NOTES:

- Aim was to combine two areas of interest for the STEM project, those being environmental science and climatology.
- This led to an idea for a potential project involving the investigation of how varying ocean conditions impact the concentration of various species of phytoplankton. Knowing what range of conditions different species can handle, coupled with the knowledge of how ocean conditions are changing due to global warming can reveal ecological and climatic insights.
- With that in mind, I chose to read this journal article, which provides an overview of the role phytoplankton play in climate and marine ecosystems and how climate change is impacting them.
- Phytoplankton refers to a broad range of photosynthetic species of viruses, bacteria, fungi, protists, animals, and archaea who are unable to move against ocean currents. As a



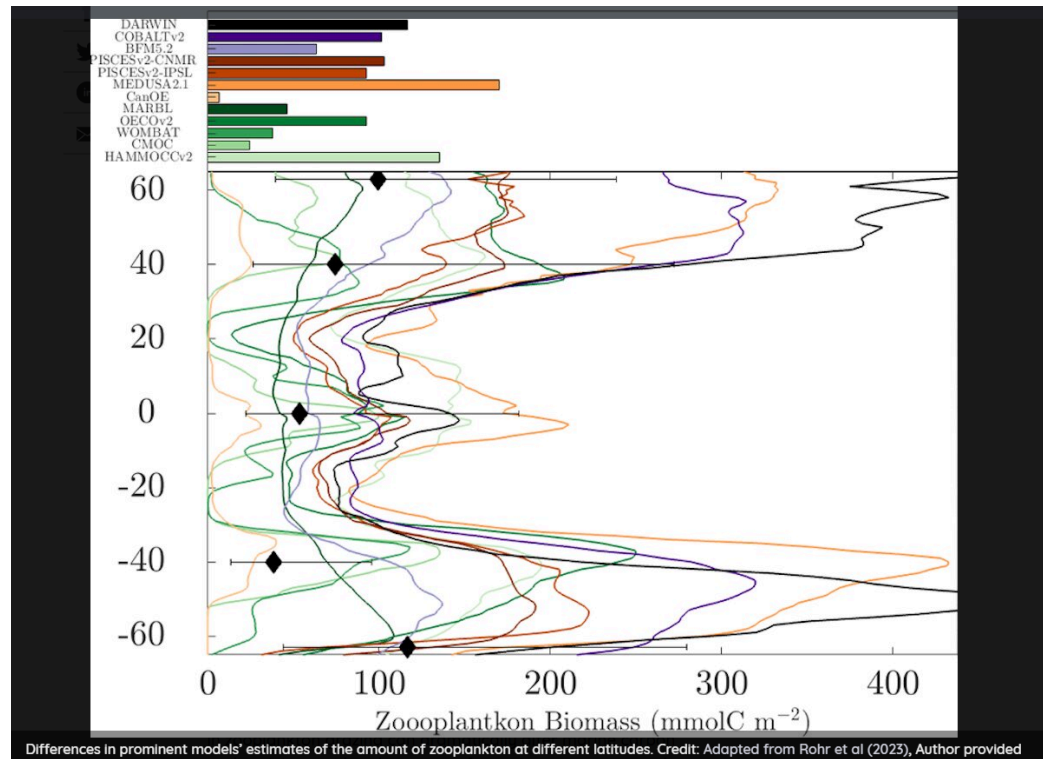
result, phytoplankton taxonomy is quite complex, and their structure, biochemical composition, habitat, and other features vary greatly.

- As photosynthetic organisms, phytoplankton are a significant source of carbon sequestration and oxygen production, accounting for 50% of global primary production. Carbon is cycled throughout the ocean through phytoplankton sinking, vertical migration of certain species, and other means (this carbon is known as oceanic dissolved organic carbon, or DOC).
- Phytoplankton also require nitrogen, phosphorus and trace metals for survival.
  - Iron in particular plays a role in increasing photosynthetic productivity.
- Phytoplankton are the base of marine food chains, serving as prey for fish, zooplankton, and various heterotrophs, and being decomposed by many bacteria and viral lysis.
- A wide range of methods exist to examine different properties of phytoplankton, including remote sensing, mass spectrometry, chromatography, and many others.
  - In particular, time series, that is, sampling of the ocean in intervals, is an important tool in studying phytoplankton in the long term. This technique has formed many other hypotheses about the ocean and revealed crucial insights about the long term trends in phytoplankton.
- Higher greenhouse gas levels have led to warmer ocean temperatures, acidification, deoxygenation, increasing contrast between freshwater and saline conditions, eutrophication, stratification, and sea level rise.
- Even without the impact of climate change, phytoplankton already adapt to seasonal changes. The article highlights numerous examples of varying seasonal changes around the globe and how that informs trends among different phytoplankton species.
- The climate-induced changes in oceans will lead to less primary production in some regions, while it will lead to greater amounts in others.
- However, shifting bloom periods and stratification may decrease the availability of certain phytoplankton species, which in turn will limit food sources for predators, impacts which will spread to the rest of the food chain.
  - Predators may travel to new regions, becoming invasive species, in order to adapt to this lack of availability, causing further disruption.
- Changing ocean conditions may also limit the availability of essential elements and trace metals.
- Another impact is the increase in harmful algal blooms, which produce toxins.
  - These toxins, such as ones produced by harmful algal blooms (HABs), travel up the food chain, eventually reaching humans. Exposure of such sea toxins to humans has devastating socioeconomic consequences.
  - HABs drain ocean resources, creating an anoxic environment.
- Conversely, phytoplankton play a significant role in climate regulation, having been a major cause of ice ages, reflecting solar radiation, and sequestering CO<sub>2</sub>.
- The impacts of climate change on the ocean and phytoplankton vary from region to region, and that making exact predictions is difficult. Hence, new and existing methods and models must be used to gain more insight.

## Article #3 Notes: Oceans absorb 30% of emissions, driven by a huge carbon pump: Tiny marine animals are key to cycle, says study

<b>Source Title</b>	Oceans absorb 30% of emissions, driven by a huge carbon pump: Tiny marine animals are key to cycle, says study
<b>Source citation (APA Format)</b>	Rohr, T., Richardson, A., & Shadwick, E. (2023, June 15). <i>Oceans absorb 30% of emissions, driven by a huge carbon pump: Tiny marine animals are key to cycle, says study</i> . Phys.org. <a href="https://phys.org/news/2023-06-oceans-absorb-emissions-driven-huge.html">https://phys.org/news/2023-06-oceans-absorb-emissions-driven-huge.html</a>
<b>Original URL</b>	<a href="https://phys.org/news/2023-06-oceans-absorb-emissions-driven-huge.html">https://phys.org/news/2023-06-oceans-absorb-emissions-driven-huge.html</a>
<b>Source type</b>	Scientific News Article
<b>Keywords</b>	No keywords are provided.
<b>#Tags</b>	<a href="#">Example of the Importance of Accurate and Precise Modeling and the Drastic Impact of Minor Changes to Model Parameters</a>
<b>Summary of key points + notes (include methodology)</b>	<p>MINI-SUMMARY:</p> <p>Modeling the carbon dioxide absorbed by phytoplankton and the resulting climate conditions has been a major challenge as different models make different assumptions about certain parameters. Namely, this article notes how different models making different assumptions about the amount of grazing pressure phytoplankton face from zooplankton leads to drastically different climate predictions, mentioning an experiment where only slightly increasing grazing pressure and phytoplankton populations led to an extra two billion tons of carbon dioxide being sequestered. As a result, it is important to more definitively determine the role zooplankton play in the carbon cycle in order to establish consistency in predictions among different models.</p>
<b>Research Question/Problem/ Need</b>	What is the exact role zooplankton play in the carbon cycle, and how can that be modelled consistently to ensure less variation in predictions about climate? More broadly, how can the carbon cycle be modeled as accurately as possible?

## Important Figures



This figure illustrates the estimates of the amounts of zooplankton present at different latitudes made by various models. Each line represents a different model. For the DARWIN and MEDUSA 2.1 models, there tends to be a higher amount of phytoplankton at all latitudes. For example, at 60 degrees, DARWIN predicts about 400  $\text{mmolC m}^{-2}$  in zooplankton biomass and MEDUSA 2.1 predicts about 325  $\text{mmolC m}^{-2}$ , whereas most other models predict an amount between 0 and 200  $\text{mmolC m}^{-2}$ . This variation is made even more apparent with the intervals placed at -60, -40, 0, 40, and 60 degrees latitude. Their presence indicates the sheer inconsistency in the amount of zooplankton that models estimate. Interestingly, given that all models estimate a lower amount of zooplankton at equatorial latitudes, it seems reasonable to conclude that zooplankton are less prevalent around the equator and more abundant at intermediate latitudes.

## VOCAB: (w/definition)

**Secondary Production-** The generation of biomass by heterotrophic organisms. In this context, this means the abundance and growth of zooplankton (in part determined by the grazing pressure on phytoplankton), and in turn, the amount of biomass available for higher-order consumers.

**Export Production-** The amount of carbon stored deep in the ocean by phytoplankton following death.

## Cited references to follow up on

University, D. (2023, June 5). *Little-known microbes could be an early warning signal of climate tipping point*. SciTechDaily.  
<https://scitechdaily.com/little-known-microbes-could-be-an-early-warning-sign>

	al-of-climate-tipping-point/
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How do interactions higher up the food chain (beyond grazing pressure from zooplankton) impact the amount of carbon dioxide that phytoplankton can sequester?</li> <li>2. Can some formula or standardized system be developed to translate real-life ocean conditions into universal parameters for different models to use?</li> <li>3. How do changing ocean conditions (acidification, higher temperatures, stratification, nutrient deficiencies, etc.) impact phytoplankton indirectly through the impacts that higher-level consumers face?</li> </ol>

## NOTES:

- This science news article takes a closer look at the marine carbon cycle.
- The ocean is a crucial component of the carbon cycle and climate, given that it contains 60 times more carbon than the atmosphere and absorbs about 30% of anthropogenic carbon emissions.
- In being a crucial carbon sink, the ocean acts as a thermostat for the climate. The amount of carbon stored in the ocean can vary, and even slight differences can result in drastic changes for the atmosphere and climate.
- Variation in carbon storage depends upon phytoplankton, as they sequester a high amount of carbon dioxide through photosynthesis, and form large carbon sinks in the deep sea following death.
- Developing proper models of the oceanic carbon cycle is crucial for having a full and accurate understanding of it and in turn its implications for the climate.
- As the climate changes, different models disagree on what will happen to primary production, the amount of carbon phytoplankton consume and consequently provide for other producers at the base of the food web, secondary production, the growth of zooplankton which impacts the fish populations who prey on them, and export production, the amount of carbon stored in the deep sea.
- The largest source of uncertainty in these models appears to be the rate at which zooplankton consume phytoplankton. Different models make different assumptions about the grazing pressures of zooplankton and the life cycles of both groups. This leads to fundamentally different predictions about carbon storage and other climate indicators when simulating various emission scenarios on different models. Until the true role of zooplankton can be properly simulated, the accuracy of these predictions cannot be guaranteed.
- Then, the article details a study where the growth rate of phytoplankton and grazing rates of zooplankton were both increased such that the carbon consumption of phytoplankton remained the same.
- Although this increase was only by a fraction of what current models assume, this resulted in an increase of 2 billion tons of annual oceanic carbon storage, and a 5 billion ton increase in the amount of carbon consumed by zooplankton. This would drastically increase the population of zooplankton for fish to feed on, which would be significant for fisheries and those who rely on fish for their diet.

- The idea is that even small variations in the role zooplankton play result in very different scenarios for the food chain and climate.
- Accurately understanding and modeling their role, therefore, is crucial for having a clearer picture of what is to actually happen with the carbon cycle, and in turn, the climate and food chain.

## Article #4 Notes: Effects of oxytetracycline on the abundance and community structure of nitrogen-fixing bacteria during cattle manure composting

<b>Source Title</b>	Effects of oxytetracycline on the abundance and community structure of nitrogen-fixing bacteria during cattle manure composting
<b>Source citation (APA Format)</b>	Sun, J., Qian, X., Gu, J., Wang, X., & Gao, H. (2016). Effects of oxytetracycline on the abundance and community structure of nitrogen-fixing bacteria during cattle manure composting. <i>Bioresource Technology</i> , 216, 801–807. <a href="https://doi.org/10.1016/j.biortech.2016.05.060">https://doi.org/10.1016/j.biortech.2016.05.060</a>
<b>Original URL</b>	<a href="https://doi.org/10.1016/j.biortech.2016.05.060">https://doi.org/10.1016/j.biortech.2016.05.060</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Composting, Denaturing Gradient Gel Electrophoresis (DGGE), Nitrogen-Fixing Bacteria, Oxytetracycline
<b>#Tags</b>	<a href="#">Definitions of Major Processes in the Nitrogen Cycle</a> <a href="#">qPCR (Quantitative Polymerase Chain Reaction) Definition</a> <a href="#">Description of DGGE (Denaturing Gradient Gel Electrophoresis)</a>
<b>Summary of key points + notes (include methodology)</b>	In order to prevent disease, the livestock industry uses veterinary antibiotics like OTC, but when using livestock manure for composting, residues are left in the soil, which can interfere with the activity of nitrogen-fixing bacteria. In this study, advanced molecular biology laboratory techniques including Quantitative Polymerase Chain Reaction (qPCR) and Denaturing Gradient Gel Electrophoresis (DGGE) were used to measure the abundance and nitrogenase (nitrogen-fixing) activity among the bacteria, while DGGE, in conjunction with calculating specialized indices and developing and analyzing a phylogenetic tree, helped to assess diversity and community organization. Findings show that OTC inhibits nitrogenase activity at a quicker rate than it does abundance, while its effects on the diversity and structure of bacterial communities remains unclear.
<b>Research Question/Problem/Need</b>	How do varying concentrations of the veterinary antibiotic oxytetracycline (OTC) in compost impact the quantity, activity, and community organization of nitrogen-fixing bacteria in the soil?
<b>Important Figures</b>	For the following figures below, CK was the control group, given 0 mg/kg of OTC. Experiment groups L, M and H were given 10, 60 and 200 mg/kg of OTC respectively.

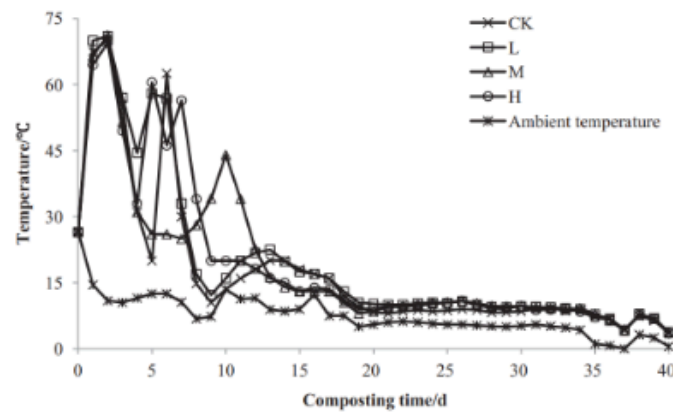


Fig. 1. Changes in temperature during cattle manure composting.

Figure 1 illustrates the temperatures for each of the piles over the course of the experiment (about 40 days). For about the first ten days, temperatures among all groups appear to oscillate rather heavily. From about days 12 to 16, the temperatures in groups CK and L are higher than those of M and H (approximately 20°C versus about 15°C). The temperature of the former groups only begin to converge with the latter ones at around day 20, with the temperature reaching around 10-12°C for all groups. Therefore, this graph indicates that higher amounts of OTC hastens temperature decline. Cooler temperatures are less ideal for most bacteria in compost piles, as they inhibit their overall activity.

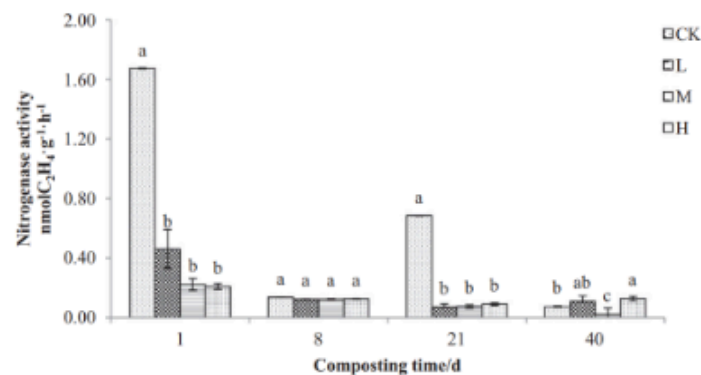


Fig. 2. Changes in nitrogenase activity during composting.

Figure 2 depicts the rates of bacterial nitrogenase activity among the four groups. These were measured on days 1, 8, 21, and 40 of the experiment. Examining these bar charts from multiple angles, it is clear that increasing OTC concentrations in compost piles inhibits nitrogen-fixing activity. On day 1, the control group has about quadruple the amount of activity in group L, and about eight times the activity of groups M and H. Even on day 8, when all groups have activity only ranging from about 0.1 to 0.2 nmol C<sub>2</sub>H<sub>4</sub>\*g<sup>-1</sup>\*h<sup>-1</sup>, the amount in the control group is slightly higher than the rest. Then, on day 21, there is a

spike of activity in the control, while activity levels in the experimental groups continue to decline. Although rates of nitrogen-fixation in L and H are greater than the control on day 40, that does not change the fact that the control group, on average, had the most nitrogenase activity over the course of the experiment. Additionally, the degree to which H and L are greater than the control on day 40 is much less than when the control was higher than all of the experimental groups on days 1 and 21. Although there are fluctuations in the values across each of the days, there is an overall decline in nitrogen-fixing activity throughout the experiment.

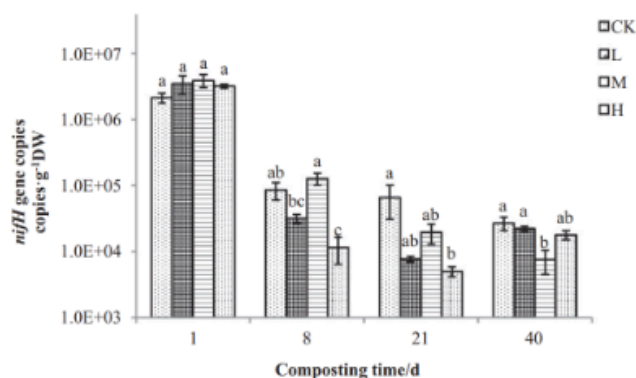


Fig. 3. Changes in *nifH* gene abundance during composting.

Figure 3 displays the abundance of nitrogen-fixing bacteria through the varying measures of *nifH* genes observed on days 1, 8, 21 and 40. While there are considerable fluctuations in *nifH* levels among each of the groups, after 40 days, the groups with the highest quantity of the genes is the control. Although not the group with the highest amount of *nifH* genes initially, the decline in *nifH* genes was lowest for the control, while the degree of decline increased the more OTC each group had. This indicates that increasing amounts of OTC decreases the quantity of nitrogen-fixing bacteria in compost piles.

#### VOCAB: (w/definition)

**Oxytetracycline** - A type of veterinary antibiotic used in the agriculture industry to inhibit pathogenic soil microbes.

***nifH* gene** - Most common marker gene (set of genes used to identify a species of an organism) used to identify nitrogen-fixing bacteria and archaea.

**qPCR** - Quantitative Polymerase Chain Reaction. A modern laboratory method used to quantitatively track gene expression, and other aspects of nucleic acids through sequencing multiple series of nucleotides.

**Nitrogenase** - Enzyme commonly found in various groups of bacteria that catalyzes the reduction of nitrogen gas (element nitrogen, N<sub>2</sub>) into ammonia (NH<sub>3</sub>), a key process in nitrogen fixation.

**Ambient Temperature** - Temperature of the environment in which experimental tools are stored.

**Ammonification** - The transformation of amine(NH<sub>2</sub>)-containing organic molecules into ammonium, NH<sub>4</sub> carried out by soil microbes.



	<p><b>Nitrification</b> - The oxidation (in this case, the addition of an oxygen atom), of ammonia and other nitrogen-containing compounds into nitrite, NO<sub>2</sub>. Nitrite is then oxidated into nitrate, NO<sub>3</sub>. This process is facilitated by microorganisms in the soil.</p> <p><b>Denitrification</b> - The removal of nitrogen from nitrogen-containing compounds. The process is the reverse of nitrification. Nitrate is reduced (meaning losing one oxygen atom) to nitrite. Nitrite is then transformed into nitric oxide, NO, and released as a gas. This part of the process is facilitated by bacteria. From there, NO turns into nitrous oxide, N<sub>2</sub>O, and ends off as nitrogen gas, N<sub>2</sub>.</p> <p><b>Nitrogen Fixation</b> - The transformation of molecular nitrogen (N<sub>2</sub>) into ammonia, NH<sub>3</sub> and similar compounds on part of nitrogen-fixing bacteria.</p>
<p><b>Cited references to follow up on</b></p>	<p>Bellenger, J.P., Xu, Y., Zhang, X., Morel, F.M.M., Kraepiel, A.M.L., 2014. Possible contribution of alternative nitrogenases to nitrogen fixation by asymbiotic N<sub>2</sub>-fixing bacteria in soils. <i>Soil Biol. Biochem.</i> 69, 413–420.</p> <p>Diaz, L.F., De Bertoldi, M., Bidlingmaier, W., 2011. <i>Compost Science and Technology</i>. Elsevier.</p> <p>Liu, D., Zhang, R., Wu, H., Xu, D., Tang, Z., Yu, G., Xu, Z., Shen, Q., 2011. Changes in biochemical and microbiological parameters during the period of rapid composting of dairy manure with rice chaff. <i>Bioresour. Technol.</i> 102 (19), 9040–9049.</p> <p>Liu, X.L., Huang, Y.M., Jiang, J.S., Huang, H., 2012. Function of microbial physiological group in carbon and nitrogen transformation during a swine manure-straw compost. <i>China J. Environ. Eng.</i> 6, 1713–1720, in Chinese.</p> <p>Zehr, J.P., Jenkins, B.D., Short, S.M., Steward, G.F., 2003. Nitrogenase gene diversity and microbial community structure: a cross-system comparison. <i>Environ. Microbiol.</i> 5, 539–554.</p>
<p><b>Follow up Questions</b></p>	<ol style="list-style-type: none"> <li>1. How do varying amounts of OTC in compost impact the carbon-nitrogen ratio in soil, and what ramifications does that have for the ecology of the soil?</li> <li>2. How does the ability to resist inhibition from OTC differ among species of nitrogen-fixing bacteria, and what factors influence that?</li> <li>3. Does increased diversity among nitrogen-fixing bacteria increase the ability to resist the hindrances of OTC?</li> <li>4. How does soil pH impact the ability of nitrogen-fixing bacteria to resist inhibition brought about by OTC?</li> </ol>

NOTES:  
Introduction:

- Oxytetracycline (OTC) is a commonly used antibiotic in the livestock industry to prevent infection. However, most livestock excrete large amounts of OTC. When these excretions, mainly manure, are used for composting, this poses a threat soil microbes, plant growth, and even human health.
- In the process of enhancing soil quality, compost that contains livestock manure is able to reduce the quantity and impact of harmful antibiotics.
- Nitrogen is an important element for life, especially when it comes to organism structure and proteins. Processes that occur along the nitrogen cycle, such as ammonification, nitrification, denitrification, and nitrogen fixation, are facilitated by nitrogen-fixing bacteria. The nitrogenase enzyme is used for nitrogen fixation specifically.
- Brief overview of methodology (see below)
- Goal of the study is identified: Determining the impact of OTC on nitrogen-fixing bacteria via examining the impact on the abundance of nifH genes (common marker gene for nitrogen-fixing bacteria) and the community structure of the bacteria.

#### Methods:

- Details how and where OTC, cattle manure and wheat straw were attained.
- Timeframe (26 Oct-5 Dec 2014) and composting parameters specified
  - Carbon/Nitrogen Ratio was 25:1, moisture content was around 55% (tap water used), dimensions and material of the composting box (foam, rectangular prism, holes for aeration, and so on), days of experiment when compost was turned, etc.
- Experimental groups: CK (No OTC), L (10 mg/kg), M (60 mg/kg), and H (200 mg/kg).
- Stratified random samples of the soil collected on days 1, 8, 21 and 40 of the experiment
- Temperature monitored via thermometer, moisture determined through drying, organic C content measured by organic carbon analyzer, and nitrogenase activity measured through incubating the sample in a flask containing acetylene ( $C_2H_2$ ) and measuring the output of ethylene ( $C_2H_4$ ) via gas chromatography
- Samples that had DNA extracted were freeze-dried, being stored at  $-20^{\circ}C$ .
- qPCR (see definition above) was then used to determine the number of nifH genes.
- Denaturing Gradient Gel Electrophoresis (DGGE) was then carried out. Specifies the amount of DNA template, various primers, water, and other pertinent fluids used (e.g. 1  $\mu L$  of DNA template, 20  $\mu L$  of water, etc.)
- For the above two processes, the sequences of nucleotides used to conduct the procedures is provided.
- The bands of the proteins from DGGE were then extracted from the electrophoresis, cloned via nucleotide sequencing, and then analyzed by computer software.
- The clusters of nitrogen-fixing bacteria were analyzed using non-weighted arithmetic means. To evaluate the diversity, specialized indices were used, including the Shannon, Simpson and Evenness indices. Other methods of statistical analysis for this study includes the Student's t, and ANOVA.

#### Results and Discussion:

- (See figure 1 above) Temperatures fluctuate greatly among the four experimental groups. All groups started out in thermophilic conditions, indicative of microbial growth and activity, and ideal composting conditions. In general, the data of the temperatures of each

group seem to indicate that OTC lowered temperature faster (which may indicate that it inhibited microbial activity).

- (see figure 2 above) Data on the nitrogenase activity among the control and three treatments clearly indicate that higher OTC levels have an irreversible, inhibitory effect on nitrogenase activity. On day 1, the control has a much higher amount of nitrogenase activity whereas all treatments have far lower ones. Due to overheating in the soil on day 8, all groups had low nitrogenase activity. However, only the control was able to bounce back, as seen on day 21.
- (see figure 3 above) Data on nifH gene abundance among the four groups illustrates that OTC reduces nifH gene levels. The article speculates that this may be a cause of the degradation of OTC into products that nitrogen-fixing bacteria have less resistance to, which ultimately leads to less bacteria being able to survive, and hence, a decreased presence of nifH genes.
- Results from the DGGE and qPCR suggest that varying levels of OTC have significant impact on the composition of nitrogen-fixing bacteria in the soil, over the course of the experiment, when comparing the control to the treatment groups, the banding patterns varied greatly.
- Calculations done using the various indices mentioned above reflect that higher OTC levels lead to less microbial diversity. However, there may be other factors at play, including the degradation of OTC, which inhibits ribosomes and tRNA. Therefore, more study is required when it comes to evaluating the impact of OTC and other antibiotics on the diversity of these soil microbes.
- The gene sequences were used to identify the bacterial species in the soil. The study speculates about what role they may play in nitrogen-fixation, as well as any resistance they may have to OTC. This is another place for future study.

#### Conclusions:

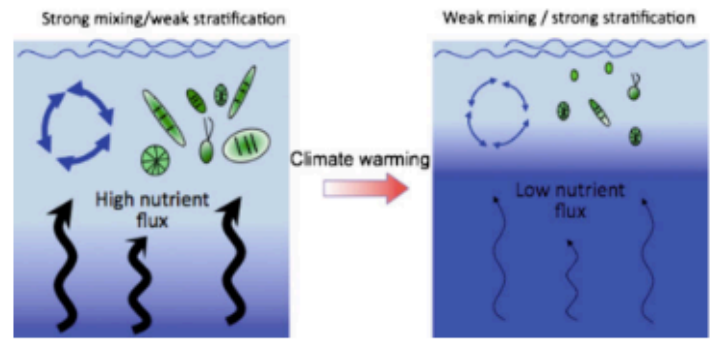
- Overall, it seems more definite that OTC has an inhibitory impact on nitrogen-fixing bacteria, reducing nitrogenase activity and gradually reducing the abundance of nifH genes. However, the impact of OTC on the diversity and community structure is less clear and needs additional study.

## Article #5 Notes: Phytoplankton response to a changing climate

<b>Source Title</b>	Phytoplankton response to a changing climate
<b>Source citation (APA Format)</b>	Winder, M., & Sommer, U. (2012). Phytoplankton response to a changing climate. <i>Developments in Hydrobiology</i> , 221, 5–16. <a href="https://doi.org/10.1007/978-94-007-5790-5_2">https://doi.org/10.1007/978-94-007-5790-5_2</a>
<b>Original URL</b>	<a href="https://doi.org/10.1007/978-94-007-5790-5_2">https://doi.org/10.1007/978-94-007-5790-5_2</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Light, Water column stratification, Temperature, Phenology, Primary production, Cell size
<b>#Tags</b>	<a href="#">Shifting Competitive Advantages for Phytoplankton</a>
<b>Summary of key points + notes (include methodology)</b>	The article begins by highlighting environmental factors that influence primary production, physiological conditions, and diversity among phytoplankton, including stratification, temperature, and light and nutrient conditions, with the consensus being that changes in these factors vary greatly among different oceanic regions, meaning that the impact on phytoplankton shall vary, as different species prefer different conditions. However, two clear trends seem to be earlier bloom periods in the spring due to warming temperatures and increased grazing pressure, as well as smaller, more buoyant species becoming more dominant, which is predicted to lessen primary production. The article concludes by highlighting the important ecological and climatic roles phytoplankton play and the drastic but not well understood ramifications the effects of climate change could have, pointing out the importance of further research and model development.
<b>Research Question/Problem/Need</b>	How will oceanic changes attributable to climate change impact the biodiversity, primary production, abundance, and distribution of phytoplankton, and how will that impact ecosystems farther up the trophic pyramid?

## Important Figures

**Fig. 1** The effect of increasing temperature on water column stratification (blue arrows), associated nutrient redistribution (black arrows), and phytoplankton production and cell size



This article uses this illustration to explain the relationship between ocean temperature and water column stratification, and what that means for the types of phytoplankton that dominate marine ecosystems. When temperatures are cooler, there is more circulation that occurs between the different levels of the ocean. This leads to more nutrients being distributed across all strata. In these conditions, larger phytoplankton species tend to thrive more. By contrast, as ocean temperatures increase due to global warming, circulation among oceanic layers decreases, and stratification occurs. This leads to lower circulation of nutrients. Smaller, more buoyant phytoplankton are predicted to thrive under these conditions.

## VOCAB: (w/definition)

**Phenology** - The study of seasonal cycles and other cyclical aspects of nature, with particular regard to plants and climate.

**Light-limited Rates of Photosynthesis** - When autotrophs are exposed to wavelengths of light that are unideal (either the frequency is too infrequent or frequent), and as a result photosynthetic output is limited by light.

**Light-Saturated Rates of Photosynthesis** - When autotrophs are exposed to ideal wavelengths of light (frequency is ideal), and as a result, some other factor other than light inhibits photosynthetic output.

**Population Size Structure** - The varying concentrations of members of different size classes.

**Turbulence** (in context of ocean) - Fluid motion that is characterized by the interactions of different currents at different magnitudes, leading to unstable velocities and pressures.

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<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How will ocean acidification impact metabolic rates among different phytoplankton species?</li> <li>2. How does the role other photosynthetic organisms play in the environment compare to phytoplankton, and will that be significant as climate change modifies the ocean?</li> <li>3. How do varying levels of competition and cooperation among different phytoplankton species influence the amount and distribution of energy transferred up the food chain?</li> <li>4. How does phytoplankton cell size impact photosynthetic output?</li> </ol>

**NOTES:****Section I: Introduction**

- Phytoplankton comprise of 50% of primary production, are base of marine food chain, and play significant role in biogeochemical cycling and climate regulation.
  - The nature in which these processes are carried out is largely based upon the composition of phytoplankton
- Therefore, a major part of understanding how oceanic conditions shall change relies on knowing on how phytoplankton shall change due to global warming, which in turn relies upon knowledge of factors that impact the ocean.
- Climate oscillations, both seasonal and long-term, have a major impact on phytoplankton conditions, as they significantly modify the abiotic and biotic conditions that impact their populations.

**Section II: Mechanistic links between climate and factors limiting primary production**

- Some of the most influential variables that impact phytoplankton include ocean temperature, stratification, prevalence of light and nutrients, play a major role in impacting phytoplankton populations
- Climate change is modifying these factors, which is to have direct and indirect effect on phytoplankton populations.
- Metabolism in primary producers is mainly limited by photosynthesis.
- Cold temperatures typically do not inhibit phytoplankton growth.
- Light-limited rates of photosynthesis are not impacted by warmer temperatures, whereas light-saturated rates of photosynthesis increase with them (makes sense considering the definitions of these two types of rates of photosynthesis described above).
- This means that in areas of light-saturated photosynthesis, blooms could increase in abundance. However, this is would lead to greater grazing rates in certain areas, altering taxonomic composition of phytoplankton.

- Stratification and the mixing of ocean layers are two opposing forces; the former occurs under warm conditions, while the latter occurs under cooler ones. Layer mixing circulates nutrients, allowing different groups of phytoplankton be exposed to the nutrients and light of different levels. However, with more stratification, nutrients, abiotic conditions become more sporadically distributed, as opposed to being uniformly the same across a given area.
- Stratification leads to the preference of lighter, smaller, more buoyant species (makes sense considering basic principles of things a kin to density)
- Stratification will lead to less nutrients in certain areas, while leading to more in other areas, as well as inhibit their upward flux. This means that the best competitors for nutrients, in addition to groups that can maintain their vertical position will thrive in these conditions.
- On the other hand, for environments that have been subject to eutrophication, it seems that phytoplankton that lie at the intermediate layers of the ocean will have the best advantage.
- Stratification may also cause deoxygenation and accumulation of other nutrients such as sulphur that are traditionally associated with eutrophication.
- The impact of climate change on marine nutrient availability varies by region.
- Light availability under changing ocean conditions will also vary by region, as stratification, reduced mixing, and earlier melting are all factors that help increase it, whereas increased winds and run-off decrease it. However, winds and run-off are regional events, and so overall, light availability, and therefore, production in phytoplankton, is anticipated to increase.

### Section III: Climate effects on phytoplankton processes

- While there are region-by-region impacts, there exist overall trends in the impacts climate change has had on phytoplankton, mainly relating to taxonomic composition, population size structure, and seasonal trends.
- Phenologically speaking, phytoplankton blooms are triggered by physiological responses to abiotic conditions, namely temperature and light. Nutrient access, predator-prey interactions, and population feedback all influence the degree at which these blooms grow. At deeper ocean layers, thermal stratification initiates blooms.
- Across multiple ecosystems, it has been observed that the time at which these blooms occur has gotten earlier, which is in agreement with the prediction of models of producer-grazer relationships.
  - This degree of advancement varies across different groups, likely due to different life strategies that different communities employ
- A possible explanation for this, based off some studies, is warmer temperatures and increased grazing pressure. These conditions are also associated with changes in taxonomic composition and population size structure.
- Changing climate conditions are also impacting the magnitude of phytoplankton blooms, but this is dependent upon light. For example, while higher intensity light increases the magnitude of blooms, when light intensity is lower, and blooms face higher temperatures and grazing pressure, the magnitude is lower.

- Impacts on photosynthetic output lack a consensus. Some regions have had higher chlorophyll outputs, while others have had lower ones.
- Smaller, more buoyant phytoplankton, being able to resist sinking, are predicted to have a competitive advantage. This includes groups such as certain cyanobacteria and flagellates.
  - Fossils and other paleoclimatological evidence support these findings.
- Moreover, in eutrophic environments, groups that are able to move upward through oceanic layers are expected to have a competitive advantage.

#### Section IV: Conclusions

- These changes are to have significant ecological and climatic implications, especially when it comes to primary production, biogeochemical cycling, and predator-prey interactions.
- With the emergence and peaks of phytoplankton blooms becoming inconsistent across different regions, this may throw off feeding patterns of zooplankton. This means that there will definitely be changes in how energy is transferred up the trophic pyramid.
- Dominance of smaller, more buoyant phytoplankton populations means less export production.
- More research into the seasonal variabilities in phytoplankton and how those are changing due to global warming is needed. A particular area of focus should be on determining how energy shall be transferred up the trophic pyramid, given these changing norms in phytoplankton.

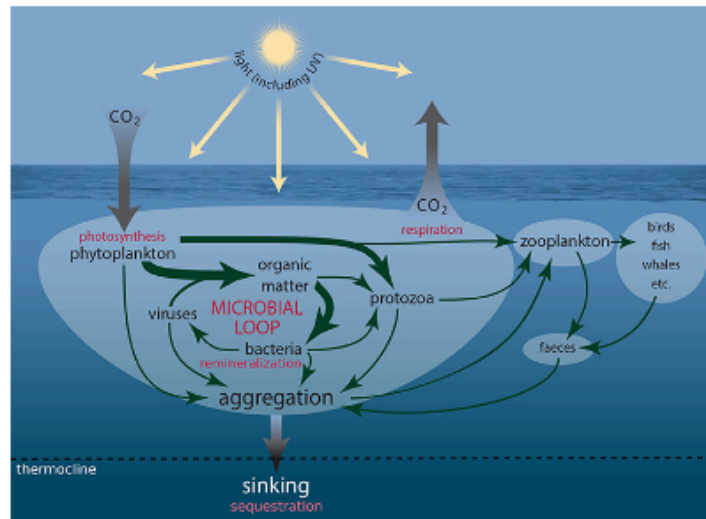


## Article #6 Notes: Southern Ocean Phytoplankton in a Changing Climate

<b>Source Title</b>	Southern Ocean Phytoplankton in a Changing Climate
<b>Source citation (APA Format)</b>	Deppeler, S. L., & Davidson, A. T. (2017). Southern Ocean Phytoplankton in a changing climate. (J. Dinasquet, Ed.). <i>Frontiers in Marine Science</i> , 4. <a href="https://doi.org/10.3389/fmars.2017.00040">https://doi.org/10.3389/fmars.2017.00040</a>
<b>Original URL</b>	<a href="https://www.frontiersin.org/articles/10.3389/fmars.2017.00040/full">https://www.frontiersin.org/articles/10.3389/fmars.2017.00040/full</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Southern Ocean, Phytoplankton, Climate Change, Primary Productivity, Antarctica
<b>#Tags</b>	<a href="#">Transferrance of Fatty Acids Up the Trophic Pyramid (focusing on cycling and dynamics of specific substances in phytoplankton)</a> <a href="#">Role of Phytoplankton in Biogenic Sulfur Budget Explanation</a>
<b>Summary of key points + notes (include methodology)</b>	This article examines how phytoplankton taxonomic composition and primary production in the Southern Ocean are being impacted by changing oceanic conditions. The Southern Ocean is broken up into five distinctive regions (mostly based on geography and oceanography). Each region's current ecological and abiotic conditions are thoroughly enumerated, and based off trends that have been seen in the data, predictions for the region are provided. Predictions, and their respective ramifications for global climate and marine ecosystems vary wildly even across adjacent regions of the same ocean. For every region, there exist multiple factors that may increase productivity, and multiple factors that may decrease it. For every region, taxonomic composition might be altered significantly, or it might not. All predicted changes are incredibly tentative. In addition to highlighting these regional complexities, the article emphasizes the need for further investigation into the rapidly changing oceanographies, while acknowledging the extreme logistical difficulties of carrying out a multi-factor experiment that would help fully understand current trends in phytoplankton dynamics.
<b>Research Question/Problem/ Need</b>	How does the impact of changing oceanic conditions due to climate change on phytoplankton and in turn marine ecosystems vary across different areas of the Southern Ocean, and what broader implications does this have for global climate?

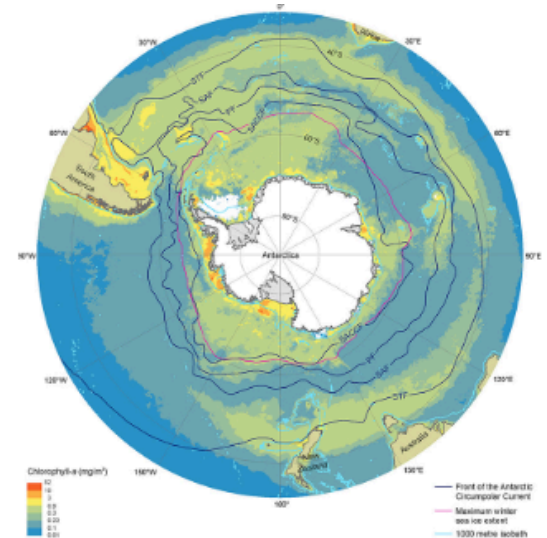
## Important Figures

FIGURE 1. SCHEMATIC SHOWING THE CONNECTIONS AMONGST MEMBERS OF THE MICROBIAL FOOD WEB AND MICROBIAL LOOP AND THE PROCESSES DRIVING CARBON TRANSFER TO HIGHER TROPHIC LEVELS AND FLUX TO THE DEEP OCEAN.



This diagram illustrates marine carbon cycling. Much of the carbon sequestered in the ocean is due to photosynthesis in phytoplankton and similar organisms. This provides a source of organic matter for bacteria, viruses and protozoa. Carbon cycling between bacteria and viruses in what is known as the microbial loop. Phytoplankton and protozoa act as the base for the rest of the aquatic food chain. The sinking of dead phytoplankton as well as fecal deposits of organisms farther up the trophic pyramid sink to deeper levels of the ocean to form carbon sinks. Carbon dioxide is released back into the atmosphere via respiration.

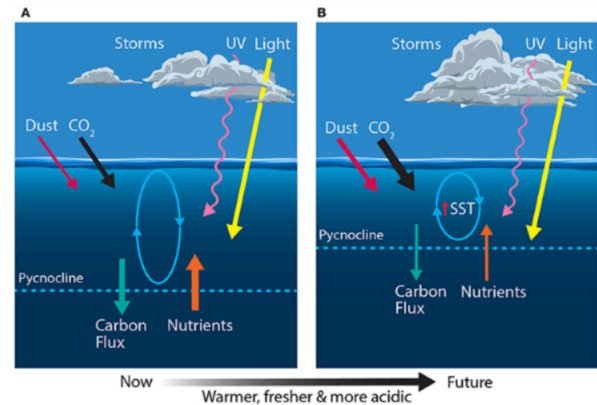
FIGURE 2. SUMMER NEAR-SURFACE CHLOROPHYLL A CONCENTRATION, FRONTAL LOCATIONS AND SEA ICE EXTENT IN THE SOUTHERN OCEAN. Chlorophyll a is determined from MODerate-resolution Imaging Spectroradiometer, Aqua satellite estimates from austral summer season between 2002/03 and 2015/16 at 9 km resolution. Black lines represent frontal positions from Orsi et al. (1995). The red line denotes the maximum extent of sea ice averaged over the 1979/80 to 2007/08 winter seasons, derived from Scanning Multichannel Microwave Radiometer and Special Sensor Microwave/Image satellite data. Light blue lines depict the 1000 m depth isobath, derived using the General Bathymetric Chart of the Oceans, version 20150318. STF, Sub-Tropical Front; SAF, Sub-Antarctic Front; PF Polar Front; SACCF, Southern Antarctic Circumpolar Current Front.



This map models the concentrations of chlorophyll A across the Southern Ocean (milligrams per cubic meter). Warmer colors indicate higher concentrations, while cooler colors indicate lower ones. Given that warmer colors seem to be present

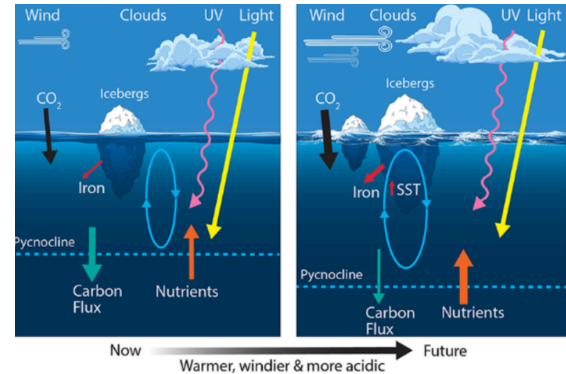
closer to coastlines of the various landmasses, and the cooler ones are farther away, there appears to be a negative association between oceanic chlorophyll concentration and distance from the coastline. This map also shows various details regarding the diminishing ice shelves and ocean currents.

FIGURE 4. SCHEMATIC SHOWING THE PRIMARY PHYSICAL CONSTRAINTS ON PHYTOPLANKTON IN THE SUB-ANTARCTIC ZONE (SAZ) (A) before and (B) after climate change, modified from [Boyd and Law \(2011\)](#). Ovals represent the depth of mixing and arrow thickness reflects relative rates of flux. SST, sea surface temperature.



This figure illustrates how ocean conditions in the SAZ are changing due to climate change. The pycnocline, the area where water density in the ocean begins increasing rapidly is predicted to move farther up the surface. More dust and  $\text{CO}_2$  exposure is expected, while the rate of circulation of nutrients, the depth of mixing, and carbon flux are expected to decrease. More storms are predicted, and sea surface temperature (SST) shall also rise.

FIGURE 6. SCHEMATIC SHOWING THE PRIMARY PHYSICAL CONSTRAINTS ON PHYTOPLANKTON IN THE PERMANENTLY OPEN OCEAN ZONE (POOZ) (A) before and (B) after climate change. Ovals represent the depth of mixing and arrow thickness reflects relative rates of flux. SST, sea surface temperature.



Another figure illustrating the impact of climate change on oceanic conditions, this time, for the POOZ region. It appears that  $\text{CO}_2$  and Fe input will increase. The pycnocline is also expected to become deeper (there appears to be a relationship between circulation and the pycnocline line) Circulation is to occur at a deeper level, and the upward flux of nutrients is predicted to increase. However, carbon storage is expected to decrease. Warmer sea temperatures, melting icebergs, windier, cloudier, stormier conditions.

The article has figures like figures 4 and 6 for other oceanic regions. While there are some unique trends predicted region by region, all figures are of a similar calibre.

The presence of unique trends among these five regions of the Southern Ocean

	<p>alone illustrates the sheer magnitude of variation present in the impact of climate change on oceanic conditions, and in turn, phytoplankton. This variation in oceanic variables warrants further study into one specific variable to see how much</p>
<p><b>VOCAB: (w/definition)</b></p>	<p><b>Marine Snow</b> - Flurries of organic material that are sinking from upper ocean layers to deeper ones.</p> <p><b>Bathymetry</b> - The measure of depth in a body of water, including but not limited to oceans, lakes, seas, and rivers.</p> <p><b>pCO<sub>2</sub></b> - The measure of partial pressure exerted by carbon dioxide gas. Used in various fields of anatomy and meteorology. Calculated by measuring the concentration of CO<sub>2</sub> in gas or other system.</p> <p><b>Pycnocline</b> - A layer within the ocean (or other water body) where density rapidly increases, i.e. where the density gradient is greatest.</p> <p><b>Antarctic Circumpolar Current</b> - A major ocean current in the Southern Ocean that flows eastward around the perimeter of Antarctica. It is the only ocean current that flows completely around the globe.</p> <p><b>Convection</b> - A large-scale movement of heat where hotter and less dense material rises while cooler, denser material sinks (with convection currents, the idea is that colder material eventually warms back up, traveling upward, before cooling and sinking again, repeating this cycle)</p> <p><b>Southern Annular Mode (SAM)</b> - Various oscillations of the westerly winds that play a significant role in determining climate in Australia, but whose variations play significant influence in climate conditions as well. It controls the north-south movement of the westerly winds, circulation in the Southern Ocean, and CO<sub>2</sub> uptake. During positive phase, winds move south towards Antarctica, and during a negative phase, move north towards the equator.</p> <p><b>Amundsen Sea Low (ASL)</b> - An area near the western coast of Antarctica characterized by its low pressure.</p> <p><b>Circumpolar Deep Water (CDW)</b> - An area of the ocean that encompasses the southern Pacific and Indian Oceans, characterized by its warm temperatures and high salinity. This is contributing to ice melt in Antarctica.</p>
<p><b>Cited references to follow up on</b></p>	<p>Arrigo, K. R., DiTullio, G. R., Dunbar, R. B., Robinson, D. H., VanWoert, M., Worthen, D. L., et al. (2000). Phytoplankton taxonomic variability in nutrient utilization and primary production in the Ross Sea. <i>J. Geophys. Res. Oceans</i> 105, 8827–8846. doi: 10.1029/1998JC000289</p> <p>Arrigo, K. R., Robinson, D. H., Worthen, D. L., Dunbar, R. B., DiTullio, G. R., VanWoert, M., et al. (1999). Phytoplankton community structure and the drawdown of nutrients and CO<sub>2</sub> in the Southern Ocean. <i>Science</i> 283, 365–367. doi: 10.1126/science.283.5400.365</p> <p>Behrenfeld, M. J., O'Malley, R. T., Siegel, D. A., McClain, C. R., Sarmiento, J. L., Feldman, G. C., et al. (2006). Climate-driven trends in contemporary ocean productivity. <i>Nature</i> 444, 752–755. doi: 10.1038/nature05317</p>

	<p>Khatiwala, S., Primeau, F., and Hall, T. (2009). Reconstruction of the history of anthropogenic CO<sub>2</sub> concentrations in the ocean. <i>Nature</i> 462, 346–349. doi: 10.1038/nature08526</p> <p>Kiene, R., Linn, L., and Bruton, J. (2000). New and important roles for DMSP in marine microbial communities. <i>J. Sea Res.</i> 43, 209–224. doi: 10.1016/S1385-1101(00)00023-X</p>
<p><b>Follow up Questions</b></p>	<ol style="list-style-type: none"> <li>1. How shall global warming-induced changes in ocean conditions impact the transference of lipids critical to reproduction up the food chain? What about other types of nutrients and substances?</li> <li>2. How shall global warming-induced changes in ocean conditions impact phytoplankton contributions to the global biogenic sulfur budget?</li> <li>3. How can computer modeling be used to emulate and then predict changes in ocean conditions and phytoplankton dynamics given drastic regional variation?</li> <li>4. How does ocean pH impact primary production in phytoplankton in oceanic waters rich in iron fertilization?</li> </ol>

NOTES:

Section I - Introduction:

- Article begins by explaining the importance phytoplankton play in cycling nutrients and acting as the base of food chains.
- Carbon cycling is mentioned (see explanation of figure 1 below), but so is the production of special sulfur compounds. Some species of phytoplankton create the compound dimethylsulfoniopropiothetin (DMSP), which when enzymatically broken down, forms dimethylsulfide (DMS). Atmospheric dimethylsulfide, when oxidized forms sulfate aerosols that reflect solar radiation. Therefore, this is another avenue through which phytoplankton play a role in climate.
- The southern ocean makes up 40% of the human-induced carbon dioxide that the ocean absorbs. If not for the southern ocean, atmospheric CO<sub>2</sub> levels would be 50% higher than they are currently.
- Changing ocean conditions due to climate (which shall impact conditions for phytoplankton) include acidification, stronger winds, greater solar radiation, rapid sea ice melting, lower salinity, changes to the distribution of nutrients, and the southward movement of certain ocean fronts.
- The article then goes on to provide a geographic definition of the Southern Ocean (SO) used in this study (everything south of the Sub-Tropical Front, which encompasses 20% of the global ocean mass). Then, based off environmental factors that influence phytoplankton, the five regions that are to be focused on in this article are defined: Sub-Antarctic Zone (SAZ), Permanently Open Ocean Zone (POOZ), Seasonal Sea Ice Zone (SSIZ), Marginal Ice Zone (MIZ), and Antarctic Continental Shelf Zone (CZ).

## Section II - Sub-Antarctic Zone:

- High nutrient, low chlorophyll region of the SO.  $p\text{CO}_2$  in this region is lower than in the atmosphere, allowing for ease of  $\text{CO}_2$  absorption. Significant contributor for the thermohaline cycle and convection currents (see definition above). Primary production in this region is limited due to various biological and physical factors, such as iron and light deficiencies, overdomination of certain species of phytoplankton that are good at absorbing silicon dioxide, among other factors. However, export of carbon to lower ocean levels is pretty high.
- Predicted changes for SAZ region include more frequent storms, increased radiation, acidity, temperatures, freshness and wind-blown dust.
- Warming ocean + more precipitation = stratification and more buoyancy needed, (as discussed in past articles)
- Less  $\text{CO}_2$  absorption as  $p\text{CO}_2$  increases
- Predicting *net results* for how phytoplankton will respond to global warming-induced ocean changes is too difficult. This is because there are multiple environmental factors that have to be considered, and they act in tandem, and modeling that with experiments is impossible. Moreover, this does not take into account interactions between phytoplankton species and with organisms farther up the food chain.

## Section III - Permanently Open Ocean Zone (POOZ):

- Natural barrier between 5-10 °C SAZ and the cold Antarctic waters (< 2 °C)
- HNLC, but to an even more extreme degree than the SAZ, although pockets of Fe from sediments
- Dominated by pico- and nano- sized flagellates.
- Antarctica Slope carries nutrients northward, however, these nutrients (e.g. nitrates, silicates, etc.) are consumed by phytoplankton, so their concentrations decreased farther northward
- Strong winter winds facilitate nutrient cycling which allows for the growth of phytoplankton in spring, although this growth is inhibited by feeding from various zooplankton species. Silicified diatoms, like in the SAZ, play the largest role in Carbon export.
- With climate change, the Westerly Winds (prevailing winds in this region) are expected to go faster, pH is decreasing, sea is becoming more fresher and warmer, cloudier conditions
- Increase in primary production from warmer temperatures are expected to be off-set by positive oscillation in SAM creating more cloud cover (i.e. less light), although, also b/c of SAM, the mixing layer is deepened, so primary production may increase
  - Deeper mixed layer = higher chlorophyll a concentration found
- Melting icebergs = more iron = more productivity and abundance for phytoplankton, more carbon exportation and carbon sequestration, higher grazing pressure,
- As climate warms, the POOZ has been moving farther and farther south, both from its northern and southern boundaries (in the north, the Polar Front has gone farther south, and in the south, more ice has melted).
- Overall, this means more area of the ocean is covered by the POOZ, which may be possible indication that primary production will increase. However, this may be complicated by changing ocean conditions
- The impacts on primary production come down to nutrients and light availability. A

deeper pycnocline would reduce light availability, but more nutrients will be available from melting icebergs, so it seems that the effects may counteract each other. Overall, it seems that blooms will become a bit more subdued.

#### Section IV - Seasonal Sea Ice Zone:

- Given that ice melts and reforms annually in this region, and the area its covers, the SSIZ is one of the largest, most dynamic and ecosystems in the world.
- Snow-covered sea ice helps regulate heat exposure in the ocean, reflecting the solar radiation while also shielding the ocean from cold atmospheric conditions in winter.
- The largest source of primary production here, are the sea ice algae, who experience very dim and cold conditions, as well as little CO<sub>2</sub> access, and high salinity, thriving best in harsh conditions.
- Primary production and biomass vary greatly even within small scales and timeframes.
- Like with all other ecosystems, the phytoplankton (in this cases embedded within the ice) is significant, being the base of the food chain in Antarctic ecosystems.
- The ice-water environment has optimal conditions (higher temp and nutrient concentrations, low salinity).
- Unoptimal conditions during the winter, with less light, but best source of food for zooplankton during this time.
- Thus any change in sea-ice conditions or phenology will have major impact for sea-ice ecosystems and phytoplankton dynamics, especially the timings of blooms.
- Interestingly, despite overall global warming trends, sea ice spread in Antarctica has slightly increased, with El Niño Southern Oscillation (ENSO) and SAM playing major part in this.
- However, ASL (see definition above) causes sea ice compression for the Amundsen and Bellinghausen Seas and expansion of the Ross Sea, so sea ice in the western Antarctic has been declining. Combined with increased impacts from global warming, ENSO and SAM will be outweighed and so sea ice will begin declining more universally.
- Sea ice expansion has reduced phytoplankton productivity. Observations of areas in SSIZ with less sea ice (later expansion, earlier retreats), have yielded little findings of increased productivity, due to light and nutrient limitations faced.
- As has been found in other articles, temporal asynchronies arise between the phytoplankton and their grazers, leading to significant changes to the distribution of energy up the trophic pyramid.
  - In particular, fatty acids significant in reproduction are being transferred up less, which can cause further constriction to population sizes up the trophic pyramid.
- Less sea ice in this region is predicted to reduce carbon exportation.
- Reduction in the contribution of phytoplankton to the biogenic sulfur budget is predicted, especially since much of the sulfur comes from the algae in the ice that is melting.
  - This includes production of Dimethylsulfoniopropionate (DMSP), which then is converted to emissions of Dimethyl sulfide (DMS) and the simpler sulfate compounds that it decomposes into which reflect solar radiation (via facilitating cloud formation).
    - Other possible uses include protecting biological tissue, controlling grazing pressure, metabolic product, antioxidant
  - Biogenic sulfur budget = the amount of sulfur produced/emitted into the

atmosphere by living things (mainly autotrophs).

- There is a higher amount of phytoplankton in thicker ice, and with the ice thinning, there are fewer amounts of them. Being concentrated in the ice, they especially serve an important role in providing food source to zooplankton. Higher up the trophic pyramid, this may lead to longer, less energy and time efficient paths for energy to travel.
- Higher precipitation (i.e. snow) is predicted. This can form depressions in the ice, flooding the area, and allowing phytoplankton on the surface to grow in these pockets, while, via reflecting more light, inhibiting growth for groups under the ice.
- Sea ice plays a significant role in regulating the carbon cycle between air and ocean. This controls pCO<sub>2</sub> and the amount of CO<sub>2</sub> absorbed between the two.
- Ice algae, whose concentrations are predicted to decline due to the loss of their melting habitats play a significant role in regulating the transferral of energy up the food chain, and with their disappearance, will cause negative consequences ecologically.

#### Section V - Marginal Ice Zone (MIZ):

- MIZ is the location where the dense sea ice transitions into open ocean.
- Region of high productivity, accounts for majority of blooms in spring and summer, however, blooms tend to be sporadic both in location and time due to low level of the mixing layer.
- Phytoplankton are trapped into ice during the winter, however, the ice has concentrated iron. So, this leads to large blooms in the spring, with high-iron, low-salinity conditions.
- Home to diverse assortment of phytoplankton species
- These large blooms contribute to large amounts of marine snow (i.e. sinking phytoplankton, CO<sub>2</sub> sequestration). The melting sea ice provides a good source of grazing and reproduction for zooplankton
- For this region, the impacts of climate change on oceans are less sea ice, more icebergs, and a positive oscillation in the SAM. The SAM currents are expected to deepen and the mixing layer and as a result decrease productivity and lead to preferences in certain species.
- Overall, predicted impact for this region is uncertain.

#### Section VI - Antarctic Continental Shelf Zone (CZ):

- High concentrations of nutrients on the surface, iron from melting continental shelf and coastal sediments, and various currents that allow for proper nutrient mixing are major sources of productivity in this region
- Blooms here are critical for terrestrial ecosystems in Antarctica.
  - Release of DMS by grazers helps indicate mating season for penguins and other benchmarks.
- Big differences between the east and west within this region

#### West Antarctica:

- Includes area around Antarctica Peninsula (Wedell Sea, Amundsen Sea)
- Warm, nutrient-filled, saline water from the circumpolar deep water (CDW) contributes significantly to ice melt but increases phytoplankton productivity, at least, theoretically. Decreasing salinity and less abundant sea ice will likely overrule CDW and other trends that increase productivity.
- Changes in this region include acidification, rapid melting in glacier mass and ice loss, greater stratification, and fresher, warmer conditions



- A major impact of freshening waters (similar to what has been noticed for other changing conditions), smaller phytoplankton cell sizes are being preferred. This has had negative impacts up the food chain (krill population decrease, preference now on scallop and less efficient fish)
- Stratification renders certain species vulnerable to the damages of excessive sunlight, so this is yet another way the increased productivity in phytoplankton is halted.
- Melting means more icebergs (but these icebergs, although more numerous, are melting rapidly), so that means more iron, which means greater productivity, as well as greater CO<sub>2</sub> sinks.
- Not much research done on the impact of pH in this area (which, remember, is measured by CO<sub>2</sub> concentration as well as pCO<sub>2</sub>)

#### East Antarctica:

- Major factors that influence productivity in this region is iron and light availability. Of the dominant phytoplankton groups, some prefer greater amounts of stratification, while others a deeper mixing layer. Phytoplankton (like everywhere else), support rest of the food chain, including the krill of this region that is a keystone species.
- Similar trends expected for this region as for what is expected in the west, although warming may be less severe here.
- With regards to acidification, polar waters are predicted to be the first to be impacted, as solubility of CO<sub>2</sub> in colder waters is increasing quickest. CO<sub>2</sub> is trapped in the water when it freezes, becoming dissolved in it, leading to acidification.
- Phytoplankton have maintained a surprising level of stability in lieu of acidification, but pH is now starting to become too low for that to continue, so taxonomic comp is changing
- Again with smaller cell sizes, there is to be less food source for grazers and less carbon export
- Multi-stressor experiments have not been performed in this region. Indeed, as we have seen throughout research hitherto, interactions between different oceanic parameters is complex to understand, but that's what's happening in the real world. This is the key area, the area not well understood currently.
  - E.g. Iron itself increases primary production, but when combined with warming, greater pCO<sub>2</sub>, and light, this creates preference for certain species
- Freshening, ice melt, warming, and stratification seem to bolster phytoplankton activity, however, freshening and acidification seem to inhibit the transfer of energy up the trophic pyramid, which could have significant negative consequences for the Antarctic food web.

#### Section VII - Conclusion:

- Understand that phytoplankton are able to withstand very extreme conditions.
- Article asserts that no clear trend primary production (or phytoplankton in general) will be discovered until 30 years from now.
- For SAZ: Surface waters are to face lower nutrient levels due to stratification. It is possible that greater iron and warmer temperatures will increase productivity, however this hinges on storms and the amount of dust they provide oceans.
- POOZ: Increased nutrient supply, mixing between layers, and melting are expected to bolster productivity, whereas limited light due to deep mixing layer, as well as cloud

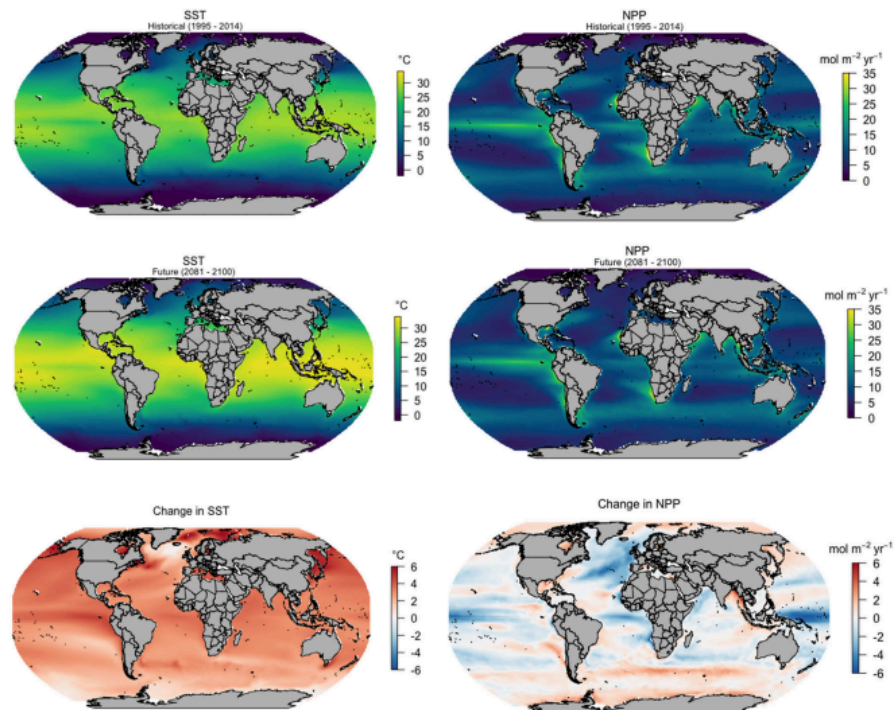
cover may limit it.

- SSIZ: With extensive melting occurring, ice algae abundance, and therefore, primary production and food availability for grazers and higher order consumers are decreasing.
- MIZ: Disruption of current phytoplankton phenology.
- CZ: Taxonomic shifts likely, especially towards smaller cell sizes; Melting, stratification are expected to generate more nutrients, thereby increasing productivity.
- Again, the key with the impacts that phytoplankton will face is the timing, interaction, magnitude, order of imposition, and other factors of the multiple stressors that phytoplankton face.
- Logistically hard to perform the multi-stressor experiments needed to generate understanding needed. Efforts in monitoring ought to be established.
- Many of the climate (and therefore oceanic) trends observed in the SO region specifically have been due depletion of the ozone layer, due to anthropogenic activity. However, many of these trends can be reversed if the ozone layer is replenished, which, currently, it seems that it is in the process of doing so.
- Another key thing to understand about phytoplankton is that many groups already face conditions that oscillate greatly, and, combined with high genetic diversity and short generation cycles, they are will fit to adapt to these conditions. However, it is unknown whether phytoplankton will be able to keep up
- Bottom line: There are to be significant changes for marine ecology, global climate, and biogeochemical cycling.

## Article #7 Notes: Monitoring and modelling marine zooplankton in a changing climate

<b>Source Title</b>	Monitoring and modelling marine zooplankton in a changing climate
<b>Source citation (APA Format)</b>	Ratnarajah, L., Abu-Alhaija, R., Atkinson, A., Batten, S., Bax, N. J., Bernard, K. S., Canonico, G., Cornils, A., Everett, J. D., Grigoratou, M., Ishak, N. H., Johns, D., Lombard, F., Muxagata, E., Ostle, C., Pitois, S., Richardson, A. J., Schmidt, K., Stemmann, L., ... Yebra, L. (2023). Monitoring and modelling marine zooplankton in a changing climate. <i>Nature Communications</i> , 14(1). <a href="https://doi.org/10.1038/s41467-023-36241-5">https://doi.org/10.1038/s41467-023-36241-5</a>
<b>Original URL</b>	<a href="https://www.nature.com/articles/s41467-023-36241-5.pdf">https://www.nature.com/articles/s41467-023-36241-5.pdf</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided.
<b>#Tags</b>	<a href="#">Use of Time Series as a Tool for Oceanic Modeling</a> <a href="#">Considering Degree of Gelatiny as a Possible Area of Study</a> <a href="#">Possible Tool of DNA Metabarcoding</a>
<b>Summary of key points + notes (include methodology)</b>	The focus of this article is twofold, focusing on the overarching trends zooplankton are undergoing due to changing ocean conditions on the one hand, and the means by which these changes can be modeled and investigated on the other. Analyzing trends from multiple time series and other long-term data on climate and ocean conditions, three broad findings include poleward migration, phenological shifts towards earlier spring and later autumn blooms, both of which impact energy transfer up and down the trophic pyramid due to asynchronization with predator and prey, along with the preference for smaller cell sizes due to higher temperatures, which is expected to reduce CO <sub>2</sub> sequestration as smaller cell size is associated with higher production of CO <sub>2</sub> as waste. However, the article points out the vast variation these trends have among different regions, highlighting the need for more research in the Southern hemisphere. To that point, the article discusses how little confidence can be placed in zooplankton models, and how better data collection methods, including DNA metabarcoding, image analysis, among other biochemical and computational methods, ought to be used to better understand the trends zooplankton are undergoing, and the ecological, biogeochemical, climatic, and societal ramifications of that.
<b>Research Question/Problem/Need</b>	How are the overall dynamics of zooplankton populations impacted by changing ocean conditions, and how can better methods in modeling said dynamics be developed?

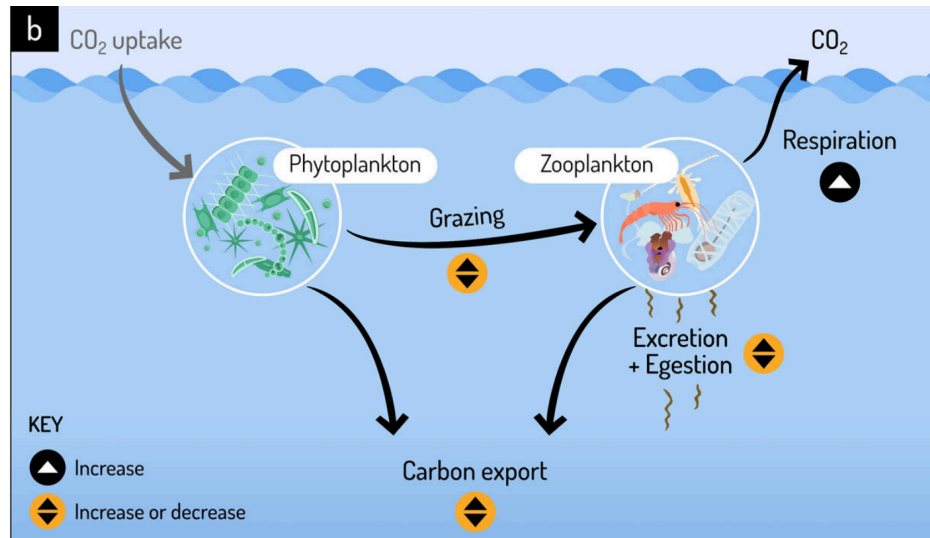
## Important Figures



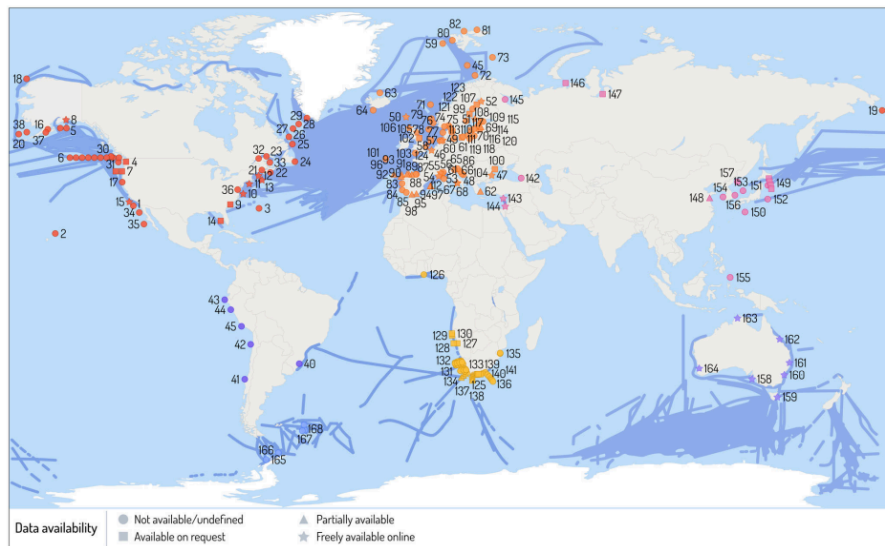
**Fig. 1 | Predicted changes in sea surface temperature (SST) and net primary productivity (NPP) for the global ocean.** Multi-model mean projections for SST and NPP from 10 CMIP6 Earth system models for the historical period (1995–2014), future (2081–2100) and the change in SST and NPP by 2081–2100 relative to 1995–2014 based on SSP5–8.5. Publicly available datasets were analysed in this

review. The 10 CMIP6 Earth system models used were ACCESS-ESM1.5, CESM2, CESM2-WACCM, CNRM-ESM2-L, GFDL-ESM4, IPSL-CM6A-LR, MIROC-ES2L, MPI-ESM1.2HR, NorESM2-LM and UKESM1-0-LL. This data can be found at: <https://esgf.llnl.gov/>.

This figure depicts past (1995–2014) ocean temperatures and amounts of primary production, provides future forecasts for said metrics (2081–2100), showing the predicted changes between time periods below. Historical data for temperature indicates equatorial waters are warmer than polar ones, with temperature of either region being approximately 25°C and between 0°C and 10°C respectively. There is a clear gradient from the equator to the poles. This holds true for the future prediction. However, the temperatures are much warmer in the predicted model. Equatorial waters are over 30°C, and polar waters have warmed up to. The mass of warm colors which graphically depict warmer temperatures has visibly expands between 1995–2014 and 2081–2100. Looking at the map of ocean temperature changes, with the exception of a few areas around Antarctica, Greenland, and Iceland, ocean temperatures increase, with the deepest shades of red, i.e. the greatest increased being concentrated in the North Pole. Meanwhile, looking at both the past and prediction models for primary production, it is difficult to easily discern any differences between the two. For both, it seems that coastal areas tend to be more productive than regions of open sea. However, the globe containing changes in production is a lot more revealing. Around the poles, the Bay of Bengal, Gulf of Mexico, Hudson Bay, and other patches of water, it is predicted that there will be light increase in primary production. However, in all other regions, primary production is expected to decline, in some places with a much faster rate than others.



This diagram is a very basic illustration of the carbon cycle, and the role zooplankton and phytoplankton play in it. What is notable about this diagram is that, with the exception of respiration, all variables have the chance of increasing or decreasing. This speaks to the overall nature of extreme variation and lack of overall trends in ocean conditions brought about by climate change.



**Fig. 3 | Map of long-term monitoring programmes for zooplankton in the global ocean.** Blue lines indicate Continuous Plankton Recorder (CPR) surveys and symbols indicate sites of specific long-term monitoring programmes (see Supplementary Data 1 for details of numbered sites). Stars indicate data is freely available to download, squares indicate data available on request, triangle indicates partially available, and circles indicate data either not available or unclear on data

availability. Only programmes where coordinates were available were plotted. Data sourced from the Marine Ecological Time Series Database, EuroSea survey and surveys undertaken as part of this review effort. More information and coordinates are provided in the Supplementary information. This figure was designed by Dr Stacey McCormack (Visual Knowledge).

This map depicts areas where zooplankton populations are studied and surveyed. There is a fairly clear Western bias here (South Africa and Japan are well-aligned with the West). The blue lines, which indicate oceanic areas where zooplankton are monitored are concentrated around the Northeast Atlantic, Southern Australian Coast, and other areas where Western presence is highly dominant. A similar pattern can be observed with the location of facilities that investigate phytoplankton. This indicates that there is dearth in many regions on of the world

	when it comes to studying and having information about zooplankton that ought to be filled (this is discussed extensively in the article).
<b>VOCAB: (w/definition)</b>	<p><b>Time Series</b> - Large amounts of data, often long-term and/or predictive in nature, that are plotted over periods of time. Presently, AI has been used to optimize this tool.</p> <p><b>North Atlantic Oscillation (NAO)</b> - Fluctuations of air pressure located in the Northern Atlantic region that influence the intensity and direction of prevailing westerly winds, and thus storm tracks in the region.</p> <p><b>DNA metabarcoding</b> - A modern, currently developing method of identifying different species of organisms from a sample of mixed-up DNA. In this process, the DNA of multiple species is augmented via PCR, and then sequenced, allowing for the organism to be identified.</p> <p><b>C:N:P Ratio</b> - The Carbon-Nitrogen-Phosphorous Ratio, also known as the Redfield ratio, is 106 to 16 to 1 ratio between the C, N and P found in phytoplankton biomass and in deep ocean waters. This equilibrium is important in maintaining the well-being of marine ecosystems.</p>
<b>Cited references to follow up on</b>	<p>Atkinson, A. et al. Questioning the role of phenology shifts and trophic mismatching in a planktonic food web. <i>Prog. Oceanogr.</i> 137, 498–512 (2015).</p> <p>Miloslavich, P. et al. Essential ocean variables for global sustained observations of biodiversity and ecosystem changes. <i>Glob. Change Biol.</i> 24, 2416–2433 (2018).</p> <p>Wright, R. M., Le Quéré, C., Buitenhuis, E. T., Pitois, S. &amp; Gibbons, M. J. Role of jellyfish in the plankton ecosystem revealed using a global ocean biogeochemical model. <i>Biogeosciences</i> 18, 1291–1320 (2021).</p> <p>Lombard, F. et al. Globally consistent quantitative observations of planktonic ecosystems. <i>Front. Mar. Sci.</i> <a href="https://doi.org/10.3389/fmars.2019.00196">https://doi.org/10.3389/fmars.2019.00196</a> (2019).</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Does a decrease in cell size in zooplankton result in a decrease in cell size for phytoplankton?</li> <li>2. Does an increase in the presence of gelatin in phytoplankton increase the rate at which carbon metabolism occurs?</li> <li>3. A common theme with shifting phenology in zooplankton is that this causes desynchronization with their predators. However, does desynchronization occur with prey (i.e. phytoplankton) too? How does zooplankton desynchronization with phytoplankton impact grazing pressure, and in turn, how does phytoplankton</li> <li>4. How can DNA metabarcoding (definition above) be used to computationally model shifting taxonomic composition in phytoplankton? How can this be connected to various oceanic conditions as parameters?</li> </ol>

## NOTES:

## Introduction/Background Information

- Zooplankton, like phytoplankton, play a significant role in the carbon pump and in marine food webs.
- Focus: Establishing oceanic changes brought about by climate change that could impact zooplankton, how zooplankton are being affected, and how these dynamics are modeled.
- When it comes to global warming-induced changes in oceanic conditions, there are broad trends, but numerous regional variations
  - Sea Surface Temperature (SST) is increasing, but at different rates in dif. regions
  - Net Primary Production (NPP) is higher towards poles, lower towards equator. Here too there is much variation; uncertainty has increased with more accurate, realistic models (CMIP5, CMIP6)

## Impact of Climate Change on Zooplankton:

- Ocean warming/acidification leads to certain groups being preferred, meaning taxonomic comp will be modified
- Use of Long-term time series for easily delineating the three key trends observed:
  - Seasonal shifts in trends, mostly earlier occurrence of spring/summer groups, and later occurrence of autumn groups
  - Geographical shift towards the poles
  - Preference of smaller sizes under warming ocean conditions
- Possible impacts on the biological carbon pump; Ecologically, the decoupling of predator-prey interactions, which would harm marine ecosystem well-being (fisheries hurt in particular)

## Seasonal Changes in Zooplankton:

- Seasonal changes vary significantly given temperature preferences and the rate of adaptations of different groups
- Highlights various examples of how zooplankton blooms and peaks are occurring much earlier
- More research has been done in the Northern Hemisphere, and there are other possible, more complex, findings in the Southern Hemisphere (i.e. no seasonal change in some regions)

## Migration:

- Poleward migration has been brought about by the need for zooplankton to maintain their optimal temperature levels
- Movement rates vary among different species, with some species not migrating poleward in any capacity, withstanding the warmer conditions, while other species are actually migrating when warmer conditions are not present
- More mobile species may be able to regulate their location a bit better, and are less sensitive to environmental conditions
- In closed systems, like the Mediterranean, instead of poleward movement, movement is towards deeper layers of ocean
- Need for further research

## Size:

- Temperature best indication of body size decline, based off the data.

- However, in southern ocean, the preference has shifted towards larger-sized species. Additionally, instances of more complex growth trends are noted.
- Other possible variables that may impact body size include species history and adaptive behaviour, competition, predation, etc.
- There are definitely connections to migration patterns and seasonal changes.

#### Climate Change & Biological Carbon Pump

- Marine heatwaves (5 days of abnormally high water temperatures), as well as events such as El Niño also heavily impact zooplankton phenology and size
  - E.g. given w/ California waters: gelatinous species preferred, disrupted predator-prey interactions
- Positive and Negative Oscillations of SAM (see definition above in article #6), and the North Atlantic Oscillation (NAO) also affect different groups of zooplankton in varying ways
- Climate change can impact zooplankton directly physiologically, and indirectly through changing their diets.
- More research needed to determine impact of other variables on phenology, size and migration patterns and the various ecological ramifications
- Zooplankton contribute to carbon exportation and cycle essential nutrients for phytoplankton and other bacteria.
  - Although, the provision of these services varies among different groups, and that is not fully understood.
- Warmer temperature leads to stratification (i.e. nutrient limitation) and higher metabolic demands, which is why smaller body sizes are preferred
- Article considers three areas in tackling how warming oceans are to impact zooplankton's role in carbon sequestration: respiration, grazing and excretion
  - Respiration: Much carbon is lost from this process. Since more CO<sub>2</sub> is released from respiration when body mass is smaller, and smaller species are being preferred, that means more carbon is being lost and sequestration is working less well. Oxygen, pH, turbulence, pressure, and other variables impact respiration.
  - Grazing Pressure: Evidence existing to support notions that grazing pressure is decreasing and increasing. Biomass and varying nutrient conditions can limit it, while warmer temperatures can expand it.
  - Excretion: Analysis of C:N:P can be measured to see if excretion will  $\pm$ . If ratio between prey and predator are similar, there is less nutrient cycling but more assimilation (which is optimal). (therefore, dif ratios means more nutrient cycling, less assimilation). Ratios are based off environmental conditions
- 2 of 3 variables are uncertain, and when layered with uncertainties about phenology, poleward shift, and size, there emerge many complexities
- In order to address this, multi-driver experiments with variables that match real-life conditions can help to develop models that can test these variables and discern trends.

#### Higher Trophic Levels:

- Zooplankton and Phytoplankton phenologies are peaking earlier at a more quicker rate than their peers up the trophic pyramid, meaning there could be negative consequences, mostly related to accessibility for fisheries (squid and crab, too)



- There exist clear links between zooplankton and fishery conditions, however, more concrete data collection is needed in this area.
- Harder to model impacts on other species higher up the food chain (e.g. whales, seabirds, seals, etc.)

#### Challenges of Modelling Zooplankton in a Changing Climate:

- Modelling is a very useful technique for modelling oceanic, phytoplankton and zooplankton dynamics. However, little confidence can be placed in some models as zooplankton are not represented or misrepresented. Three components needed (play biggest role in influencing biogeochem cycles– thus getting parameters for models):
  - Rates (things liked respiration, excretion, ingestion, etc.)
  - Traits (size, reproduction, stoich)
  - Stocks (biomass, abundance, area of coverage)
- Quantitative information on individual zooplankton species is needed. Use of DNA metabarcoding and other new, advanced technique to add to data deficit.
- Not enough empirical data to compare models to the real world conditions to carry out validation.
- Challenges can be overcome with image analysis and related advancements, especially when it comes to biomass estimation and assessing tax comp.
- Lack of data on zooplankton that live at lower depths of the ocean, which leads to information about nutrient cycling and other important processes being ignored.
- Integrating zooplankton into modelling systems can help address a myriad of experimental questions that were previously unanswerable. Progress has been made, but more needs to be made in order to get a fuller picture for impact of zooplankton.
  - This observation of inadequate data on zooplankton aligns a lot with how in Article #3, their role in carbon cycle was not well understood.

#### Sustained Observations Quantifying Impacts of Climate Change

- Article discusses the benefits of long-term ecological/climatic studies (provides insights over a longer period of time, allowing for more trends to be found, changes to be assessed, etc.) - important modeling crucial biological, biogeochemical, and physical phenomena
- The Longterm Ecological Research Network (LTER)
- Of 168 Long-Term Zooplankton monitoring programs, 81% is partially or completely unavailable. Severe lack of accessibility inhibits ability for scientific study to be carried out.
- Highlights the examples of past successes of experiments using LTER programmes to make a case for expanding accessibility.
- There also many opportunities for groups of zooplankton scientists worldwide to collaborate. With many gaps in data in certain regions of the ocean, article calls for these regions to be investigated more deeply.

#### The Future of Global Zooplankton Research

- So the main idea is that seasonality, pole-bound migration, and preferences for smaller body sizes are the 3 most definitive trends, which can have significant implications for biogeochem cycling and energy transferral up the trophic pyramid

- Shift from nets to sensors, methods involving nucleic acids, image tech, time series methods. Combining the best of old and new methods → collect best data → use that to create strong conclusions (use by policymakers and other members of society)
- Variety of sampling methods ought to be used. Best one for the given context.
  - Using satellites to connect zooplankton dynamics with oceanic/climate conditions
- Collaboration among different fields of scientists is necessary to attain data, offer methods of data collection, and attainment of necessary parameters.
  - This is what will lead to the multi-drive experimental models that can be used to test multiple experiments very easy through the use of computer modeling!

#### Linking Zooplankton Observations To Global Needs

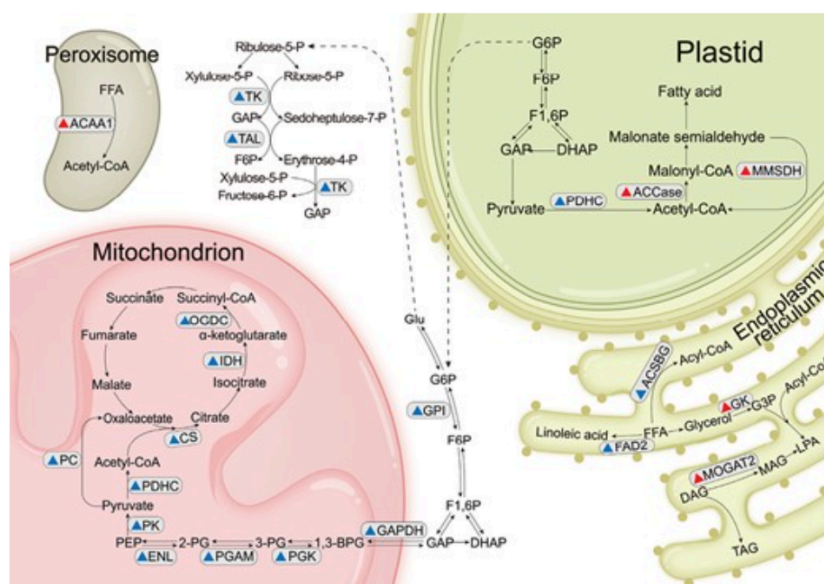
- Modeling zooplankton dynamics will help humanity understand and protect them and the ocean better, helping environment in general
- Article identifies 4 goals going forward w/ regards to zooplankton
  - Create new time series programmes in addition to protecting existing ones
  - Improve integration of time series data
  - Better understand the impact of climate change on zooplankton
  - Make use of collaborative, interdisciplinary approaches in research
- That way, zooplankton can be better understood. Ramifications from the research can be developed, and society assisted.

## Article #8 Notes: DNA methylation and gene transcription act cooperatively in driving the adaptation of a marine diatom to global change

<b>Source Title</b>	DNA methylation and gene transcription act cooperatively in driving the adaptation of a marine diatom to global change
<b>Source citation (APA Format)</b>	Wan, J., Zhou, Y., Beardall, J., Raven, J. A., Lin, J., Huang, J., Lu, Y., Liang, S., Ye, M., Xiao, M., Zhao, J., Dai, X., Xia, J., & Jin, P. (2023). DNA methylation and gene transcription act cooperatively in driving the adaptation of a marine diatom to global change. <i>Journal of Experimental Botany</i> , 74(14), 4259–4276. <a href="https://doi.org/10.1093/jxb/erad150">https://doi.org/10.1093/jxb/erad150</a>
<b>Original URL</b>	<a href="https://doi.org/10.1093/jxb/erad150">https://doi.org/10.1093/jxb/erad150</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Adaptation, diatom, DNA methylation, global change, high CO <sub>2</sub> , transcriptomics, warming
<b>#Tags</b>	<a href="#">Metabolism of Various Macromolecules</a>
<b>Summary of key points + notes (include methodology)</b>	This study aims to illustrate how, contrary to current beliefs, DNA methylation can facilitate, not just inhibit gene expression, and how phytoplankton may use this hypothesized genetic relationship to adapt to changing ocean conditions. To investigate possible adaptive capabilities induced by DNA methylation, model organism <i>Phaeodactylum tricornutum</i> was stored in four treatments including one with ambient temperature and CO <sub>2</sub> (used for pH) i.e. the control, one with higher temperatures, one higher CO <sub>2</sub> levels, and one treatment that combined those two conditions for a duration of two years. DNA and RNA of samples were extracted and sequenced in order to quantify gene expression (FPKM) and the rate of DNA methylation respectively. Based off regulations of the differentially expressed genes (DEGs) observed, it appears that warmer and more acidic oceans will inhibit carbon and amino acid metabolism, thereby necessitating an increase in fatty acid metabolism. Although many of the DEGs that create these conditions were part of DNA that was methylated, this was only the case for approximately 20% of the time. As such, DNA methylation cannot be definitively declared as an adaptive method for phytoplankton, but there appears to be evidence that it can work in concert with gene expression to bring about adaptation. Overall, future areas of focus include more direct impacts these findings have on phytoplankton, and the molecular mechanics behind DNA methylation and other relevant biochemical processes.
<b>Research Question/Problem/Need</b>	Can DNA methylation facilitate gene expression, and how can that act as a tool for adaptation of phytoplankton to warmer, more acidic ocean conditions?

## Important Figures

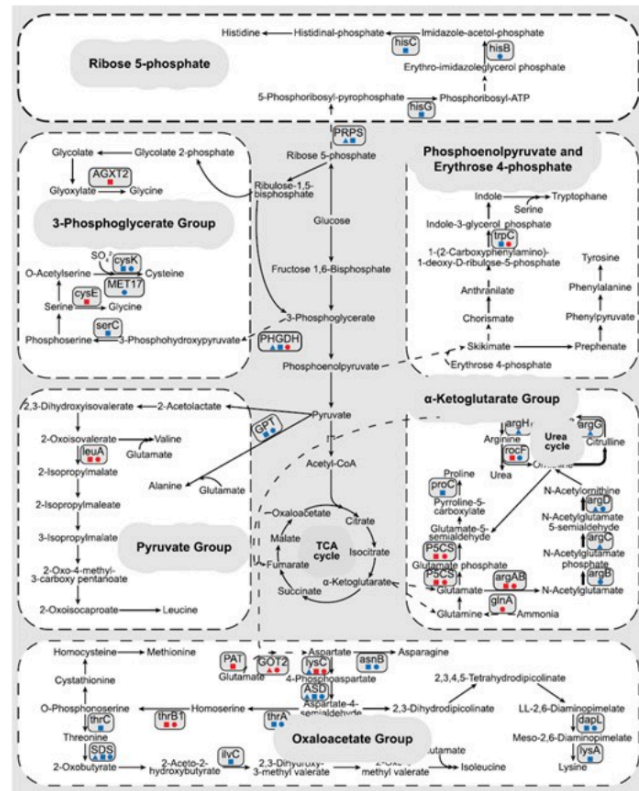
Fig. 1.



Down-regulation of central carbon metabolism and up-regulation of fatty acid metabolism after long-term high  $\text{CO}_2$  adaptation. Schematic representation of proposed metabolic pathways in long-term high  $\text{CO}_2$ -adapted *Phaeodactylum tricornutum* cells, including glycolysis, the OPPP, TCA cycle, fatty acid metabolism, and triacylglycerol (TAG) accumulation. Red or blue triangles represent the genes that were significantly up- or down-regulated ( $\text{FDR} < 0.05$  and  $|\log_2\text{FC}| > 1$ , see the Materials and methods), respectively, after long-term high  $\text{CO}_2$  adaptation. Solid arrows represent a one-step reaction, and dashed arrows showed multi-step reactions. The metabolic genes are shown in different organelles according to protein localizations predicted in the literature. Glu, glucose; G6P, glucose-6-phosphate; F6P, fructose 6-phosphate; F1,6P, fructose 1,6-bisphosphate; GAP, glyceraldehyde 3-phosphate; DHAP, dihydroxy acetone phosphate; 1,3-BPG, 1,3-bisphosphoglycerate; 3-PG, 3-phosphoglycerate; 2-PG, 2-phosphoglycerate; PEP, phosphoenolpyruvate; ribulose-5-P, ribulose 5-phosphate; xylulose-5-P, xylulose-5-phosphate; ribose-5-P, ribose-5-phosphate; sedoheptulose-7-P, sedoheptulose 7-phosphate; erythrose-4-P, erythrose 4-phosphate; G3P, glycerol-3-phosphate; LPA, lysophosphatidic acid; MAG, monoacylglycerol; DAG, diacylglycerol; TAG, triacylglycerol; GPI, glucose-6-phosphate isomerase; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; PGK, phosphoglycerate kinase; PGAM, 2,3-bisphosphoglycerate-dependent phosphoglycerate mutase; ENL, enolase; PK, pyruvate kinase; PDHC, pyruvate dehydrogenase complex; PC, pyruvate carboxylase; CS, citrate synthetase; IDH, isocitrate dehydrogenase; OGDC, oxoglutarate dehydrogenase complex; TK, transketolase; TAL, transaldolase; ACCase, acetyl-CoA carboxylase; MMSDH, malonate-semialdehyde dehydrogenase (acetylating); ACSBG, long-chain-fatty-acid-CoA ligase; FAD2, omega-6 fatty acid desaturase; GK, glycerol kinase; MOGAT2, acylglycerol O-acyltransferase 2; ACAA1, acetyl-coenzyme acyltransferase.

Figure 1 - This figure is a diagram including the various reactions and metabolic pathways of carbon metabolism. Blue triangles indicate downregulated genes for the specific chemical reaction/enzyme of the metabolic process, while red triangles indicate upregulation. There are significantly more blue triangles in the diagram, indicating that most respiration processes are being inhibited. In particular, all mitochondrial processes are to be inhibited under high  $\text{CO}_2$  (pH) conditions for phytoplankton.

Fig. 2.

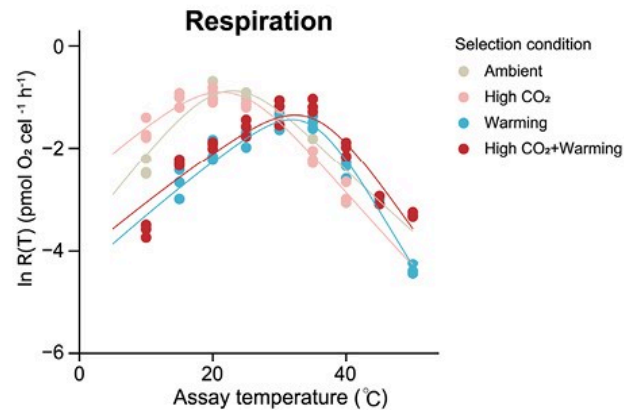


Potential changes of amino acid metabolic pathways after high CO<sub>2</sub> and/or warming adaptation. Triangles, squares, and circles represent the significantly expressed genes (FDR <0.05 and |log<sub>2</sub>FC| >1, see the Materials and methods) after long-term high CO<sub>2</sub>, warming, and high CO<sub>2</sub>+warming adaptation compared with that under ambient condition, respectively. The red and blue colour in a certain symbol represent significantly up- and down-regulated genes, respectively. Grey indicates that genes were not significantly regulated. PAT, aspartate-prephenate aminotransferase; GOT2, aspartate aminotransferase; asnB, asparagine synthase; lysC, aspartate kinase; ASD, aspartate-semialdehyde dehydrogenase; thrA, threonine synthase; thrB1, threonine kinase; thrC, threonine synthase; SDS, L-threonine ammonia-lyase; ilvC, ketol-acid reductoisomerase; dapL, L, L-diaminopimelate aminotransferase; lysA, diaminopimelate decarboxylase; glnA, glutamine synthetase; argAB, amino-acid *N*-acetyltransferase; argB, acetylglutamate kinase; argC, *N*-acetyl-γ-glutamyl-phosphate reductase; P5CS, Delta-1-pyrroline-5-carboxylate synthetase; argD, acetylmornithine aminotransferase; argG, argininosuccinate synthase; argH, argininosuccinate lyase; rocF, arginase; proC, pyrroline-5-carboxylate reductase; trpC, indole-3-glycerol phosphate synthase; PRPS, ribose-phosphate pyrophosphokinase; hisG, ATP phosphoribosyltransferase; hisB, imidazoleglycerol-phosphate dehydratase; hisC, histidinol-phosphate aminotransferase; PHGDH, D-3-phosphoglycerate dehydrogenase; serC, phosphoserine aminotransferase; cysE, serine *O*-acetyltransferase; MET17, *O*-acetylhomoserine; cysK, cysteine synthase; AGXT2, alanine-glyoxylate transaminase; GPT, alanine transaminase; leuA, 2-isopropylmalate synthase.

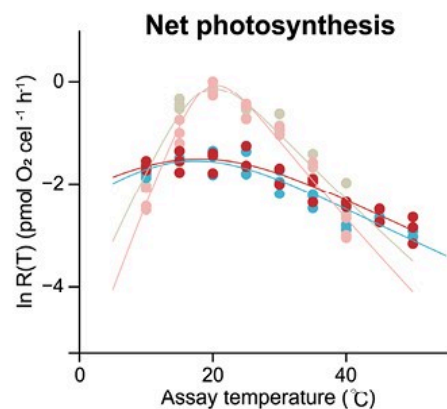
Figure 2 - This figure illustrates the various possible manners in which amino acid metabolism may be inhibited by the inhibition of carbon metabolism. Various pathways, processes, and reactions are included. Triangles, squares and circles represent the high CO<sub>2</sub>, high temperature, and high CO<sub>2</sub> and temperature treatments, respectively. Among triangles, there are 9 that are blue, and 1 that is red. Among squares, there are 17 that are blue and 10 that are red. Among circles, there are 12 that are blue, and 10 that are red. This indicates that amino acid metabolism processes inhibited are most numerous for the phytoplankton exposed to high temperature conditions (the article does indeed discuss this

finding the subsequent section), However, along with the high CO<sub>2</sub> and temperature treatments, they have the highest amount of genes in the amino acid metabolism process that are being upregulated. This is unlike with high CO<sub>2</sub> treatment, which, despite having the lowest amount of downregulated genes, has the least upregulated ones. Although, this may be an indication that this treatment group has a weaker impact either way on amino acid metabolism.

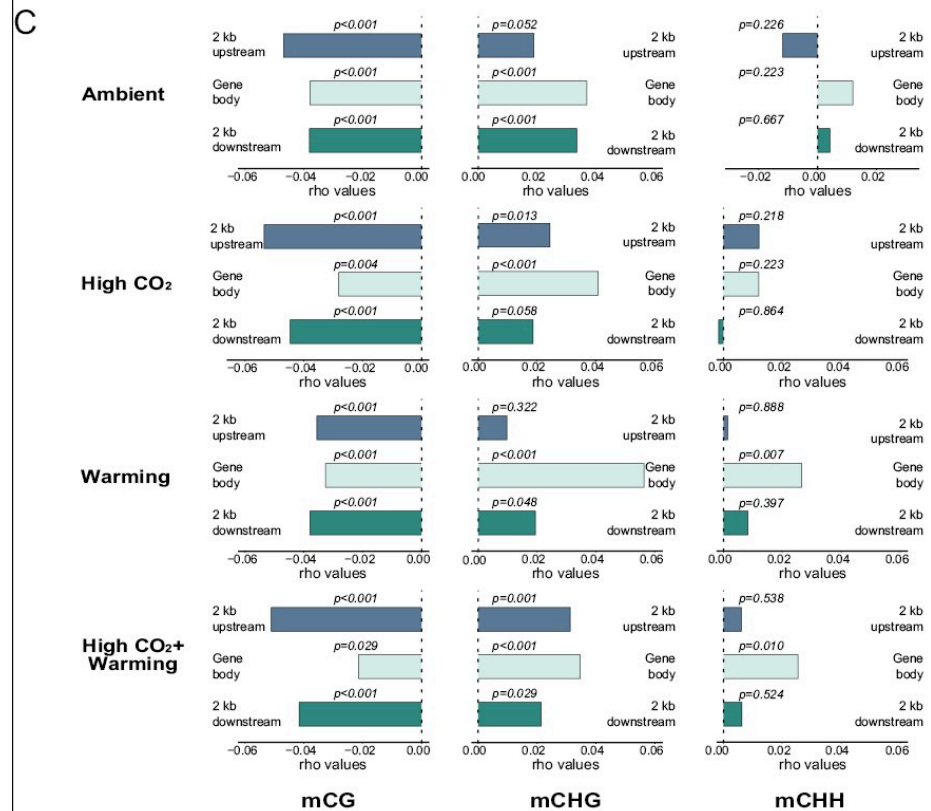
**B**



**C**



This figure displays the rates of net photosynthesis and respiration among the four treatments given assay temperature. Evidently, for all treatments of both metrics, there is a parabolic relationship that concaves down. For respiration, the maximum points of the parabola are just above -2 pmol O<sub>2</sub> per cell per hour for the warming and combined treatment, whereas it is 0 for the other two treatments. This lines up with the finding that photosynthesis is inhibited most under warmer temperature conditions, and not so much under high CO<sub>2</sub> conditions. For respiration, the peaks for the warming and combined groups are lot farther up in temperature (along x-axis) than non-heated treatments, around 35°C, while non-heated treatments reach their optimum respiration rate at 20°C. The peak respiration of the latter is slightly higher, at around -1 pmol O<sub>2</sub> per cell per hour.



Graphical depiction of the correlations between various types of DNAMs and gene expression within 2kb either direction of the gene body. There are 3 types of DNAM investigated, mCG, mCHG, and mCHH. Overall, it appears that for mCG, there is a statistically significant negative correlation with gene expression. For mCHG, all but one (assuming  $\alpha = 0.05$ ) have a statistically significant positive correlation with gene expression, while for mCHH, there are few statistically significant regressions to observe any coherent correlation with gene expression.

#### VOCAB: (w/definition)

**DNA Methylation** - The placement of a methyl group, functional group  $\text{CH}_3$ , on the 5-Carbon of the carbon ring of the cytosine nitrogenous base. This serves as a means of gene regulation. The methyl groups attracts proteins that repress gene expression and prevent the binding of transcribing factors (the proteins involved in converting RNA in to DNA)

- Destroy mRNA post-transcription
- Inhibition of promoters

**Epigenetics** - Changes in gene expression induced by the environment (NOT a change in the genome itself)

**Fragment per Kilobase of Transcript per million Mapped Reads (FPKM)** - Means by which gene expression level is normalized, measured. Done by based on gene length and total number of mapped reads.

**Differentially Expressed Gene (DEG)** - Statistically significant difference between the levels of gene expression between two groups.

	<p><b>False Discovery Rate (FDR)</b> - The rate at which the alpha level is exceeded among multiple significance tests. (similar to singular alpha level, to be statistically significant, FDR must be &lt;0.05 as per industry standards).</p> <p><b>Signal Transduction</b> - The production of a physical or chemical transmission that travels from the exterior to the interior of the cells via a series of molecular events.</p> <p><b>Downregulation</b> - When gene regulation is performed such that more of a protein or other biochemical compound is produced less. (i.e. gene expression is decreased)</p> <p><b>Upregulation</b> - When gene regulation is performed such that more of a protein or other biochemical compound is produced more. (i.e. gene expression is increased)</p> <p><b>Glycolysis</b> - Metabolic process involving ten enzyme-facilitated chemical reactions that transform glucose into pyruvate. This occurs in the cytoplasm of the cell for most organisms.</p> <p><b>Oxidative Pentose phosphate pathway</b> - A metabolic pathway involved in the respiration process. Comprised of the oxidative phase which produces NADPH, a molecule cycled through (but not directly involved in or being changed by) various anabolic pathways, and the non-oxidative phase where 5-carbon sugars are produced.</p> <p><b>Citric Acid Cycle (aka Krebs Cycle, many names)</b> - Major process in cellular respiration. Through it, CO<sub>2</sub> waste product is released, Acetyl-CoA is oxidized, producing compounds that are then used in the next part of the respiration, the electron transport chain, where ATP is produced.</p> <p><b>Porphyryns</b> - A broad umbrella terms that refers to nitrogenous, water-soluble pigments</p>
<p><b>Cited references to follow up on</b></p>	<p>Hutchins DA, Walworth NG, Webb EA, Saito MA, Moran D, McIlvin MR, Gale J, Fu F. 2015. Irreversibly increased nitrogen fixation in <i>Trichodesmium</i> experimentally adapted to elevated carbon dioxide. <i>Nature Communications</i> 6, 1–7. <a href="https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1038/ncomms9155">https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1038/ncomms9155</a>.</p> <p>Ferrari M, Torelli A, Marieschi M, Cozza R. 2020. Role of DNA methylation in the chromium tolerance of <i>Scenedesmus acutus</i> (Chlorophyceae) and its impact on the sulfate pathway regulation. <i>Plant Science</i> 301, 110680. doi:10.1016/j.plantsci.2020.1.</p> <p>Jiang H, Gao K. 2004. Effects of lowering temperature during culture on the production of polyunsaturated fatty acids in the marine diatom <i>Phaeodactylum tricorutum</i> (Bacillariophyceae). <i>Journal of Phycology</i> 40, 651–654. <a href="https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1111/j.1529-8817.2004.03112.x">https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1111/j.1529-8817.2004.03112.x</a>.</p> <p>Jin P, Gao K, Beardall J. 2013. Evolutionary responses of a coccolithophorid <i>Gephyrocapsa oceanica</i> to ocean acidification. <i>Evolution</i> 67, 1869–1878.</p>



	<a href="https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1111/evo.12112">https://doi-org.ezpv7-web-p-u01.wpi.edu/10.1111/evo.12112</a> .
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Does an increase in the concentration of phosphate and other phosphorous-containing biological compounds increase the rate of amino acid metabolism in phytoplankton? <ul style="list-style-type: none"> <li>- Possible connections to the Redfield Ratio?</li> </ul> </li> <li>2. How does a decrease in carbon metabolism in phytoplankton impact to transfer of crucial fatty acids up the trophic pyramid?</li> <li>3. How does iron fertilization differentially impact the rates of catabolism and anabolism of amino acids? How does this change given different concentrations of iron?</li> <li>4. How do diversions from the Redfield Ratio impact the range of light phytoplankton are able to absorb and use as energy during photosynthesis?</li> </ol>

## NOTES:

## Abstract:

- DNA Methylation is known for its largely regulatory functions, but an area that has yet to be investigated is the potential positive interactions it could have with gene transcription.
- Brief discussion of methodology: Various experimental groups of *Phaeodactylum tricornutum* (hereafter *P. tricornutum*) were exposed to high temperatures and CO<sub>2</sub> levels, with analysis of transcription and phenotypes and bisulfite sequencing being performed.
- Findings show that experimental groups with higher CO<sub>2</sub>, or groups with CO<sub>2</sub> and warm temperatures that had the DNA methylation expressed more genes.
  - Article refers to this as DEG, differentially expressed genes, and DMR, differentially methylated regions
- These DEGS then acted in concert with DNA methylation to regulate processes including metabolism of carbon, amino acids, and lipids, production of ribosomes, recycling denatured proteins, among others.
- This illustrates the capabilities of phytoplankton to adapt to changing ocean conditions.

## Introduction:

- DNA Methylation (hereafter DNAM) is an epigenetic process, meaning it regulates the gene expression but not composition. Short-term modifications can lead to long-term adaptations across a species (via the means of evolution, natural selection, etc.)
- Area that has not been previously focused on is the impact of DNAM on long term adaptations in phytoplankton, especially given changing ocean conditions.
- DNAM theorized to play major role in phytoplankton evolution due to findings in previous research that DNAM regulated phenotypic traits under high CO<sub>2</sub> conditions
- Aim is to investigate possible positive relationship between DNAM and transcription.
- Model species *P. tricornutum*. Genes have been sequenced, mutations quantified, and methylation information has been obtained. There are many genetic and epigenetic factors at play that need to be investigated (namely, are changes observed due to environmental conditions or biochemical processes?)

- Four treatments: Control (400  $\mu\text{atm}$  of  $\text{CO}_2$ , 15  $^\circ\text{C}$ ); High  $\text{CO}_2$  (1000  $\mu\text{atm}$  of  $\text{CO}_2$ , 15  $^\circ\text{C}$ ); High Temperatures (400  $\mu\text{atm}$  of  $\text{CO}_2$ , 20 $^\circ\text{C}$ ); Both High  $\text{CO}_2$  and Temperature (1000  $\mu\text{atm}$  of  $\text{CO}_2$ , 20 $^\circ\text{C}$ )
- *P. tricornutum* grown under respective treatments for approximately two years.
- After this period of time, transcription analysis, bisulfite sequencing. DNAM and gene expression were correlated and then used to consider possible biological processes subject to regulation.
- Important insights for evolutionary biology of phytoplankton.

#### Methodology:

##### Conditions:

- Describes/justifies the use of the four treatments mentioned above. (1000  $\mu\text{atm}$   $\text{CO}_2$  and 20 $^\circ\text{C}$  temperature are the projected  $\text{CO}_2$  and temperature levels by end of 21st century in high emissions scenario, while controls were chosen given ideal conditions for model organism)
- 100  $\mu\text{mol}$  flux of photons in a 12 hour light and dark cycle
- Cloning performed in order to create enough specimens for all four treatments
- For  $\text{CO}_2$ , margin of error of 30  $\mu\text{atm}$  for the control, 50 for the experimental groups.
- Study describes how various aeration and atmospheric regulation techniques were used to maintain desired  $\text{CO}_2$  levels. Use of bottles to prevent undesirable air exchange.
- Cellular density was kept at constant  $4 \times 10^4$  and  $5 \times 10^5$  cells per ml, regulated as needed every 5-7 days
- About 1600 to 1700 generations were produced over the course of the two-year experiment.

##### Qualifying Phenotypic Traits:

- Main traits investigated include net photosynthesis and respiration.
- Samples of phytoplankton were placed in 5 mL Tris-buffered solutions that were adjusted to match  $\text{CO}_2$  and pH conditions of their respective treatments. 10 minute acclimation period
- Oxygen evolution in the light and oxygen consumption in the dark were measured using oxygen electrodes in order to measure net photosynthesis and respiration. While this is standard methodology, rates may vary due to varying photosynthetic quotient in phytoplankton and macromolecular composition. Either way, both of these metrics were normalized.
- Modeling net photosynthesis in relation to temperature was done using the Sharpe-Schoolfield equation (includes variables such as rates of metabolism, metabolic activation energy, temperature at which enzymes are inhibited, temperature in kelvin, extensive use of natural logarithms)
  - An equation relating the highest metabolic rates to their optimum temperature levels was also used.

##### Analysis of Transcription

- Specimen from all four treatments were removed from their respective environments at the same point in the cell cycle, during light exposure, for the purposes of RNA extraction, being stored in liquid nitrogen as -80 $^\circ\text{C}$

- Extracted via the use of Trizol reagent kit, following manufacturing instructions, quality assessed by using the software Agilent 2100 Bioanalyzer and performing electrophoresis.
- Enrichment of RNA (excising of extraneous sequences) was performed using oligosaccharides for eukaryotic cells, and prokaryotic cells using a magnetic kit
- mRNA was then reverse-transcribed into cDNA (c = copied/complimentary). The second strand of DNA was then created using DNA polymerase and other enzymes, with ligation process (i.e. connection of Okazaki fragments) also occurring
- 12 samples for each treatment (of sample size of about 40 million, that is reading of DNA sequences. Due to quality control to maximize data quality and accuracy, only about 94% of sequences were read)
- This DNA was then matched to *P. tricornutum* ribosome database to determine which of the reads were valid (about 90 to 95%).
- Gene expression quantified by calculating the fragment per kilobase of transcript per million mapped reads (FPKM)
- DNA was determined to be a DEG by analyzing the differential RNA expression between two different groups and finding genes with a false discovery rate (FDR) of less than 0.05, and that had absolute fold changes.

#### Gene Mapping

- All of these genes were then mapped on the Gene Ontology (GO) software, where the DNA was analyzed and assigned to one of three categories (this is known as annotation): molecular function, biological process, and cellular component
- Metabolic and signal transduction pathways were examined using Kyoto Encyclopedia of Genes and Genomes (KEGG)

#### Analysis of DNAM

- Similar to transcriptional analysis, specimens from all four treatments were removed from their respective environments at the same point in the cell cycle, during light exposure, to analyze the DNAM
- DNA was extracted, its efficacy verified through electrophoresis, and its concentration measured through a spectrophotometer.
- In order to perform the bisulfite sequencing, DNA library was prepared. DNA was split up into multiple fragments, and adenine was added to the 3' prime end of the strand. All DNA was then ligated.
- During the bisulfite treatment, non-methylated DNA was converted into uracil. Meanwhile, methylated DNA was able to be converted successfully by the bisulfite.
  - (Answers question of why use bisulfite – good for differentiating between methylated and non-methylated DNA)
- For the DNA that was converted, the DNA was augmented using PCR, and then sequenced. However, raw reads were eliminated from analysis if over 10% of nucleotides were unknown and/or 40% of low quality nitrogenous bases had to be removed.
- BSMAP software was used to map the clean reads of DNA to *P. tricornutum* database. This software is able to detect methylation patterns. Using this, methylation rate (amount of methylation that occurred throughout entire genome

sample, each chromosome, and each region) was calculated. Average methylation rate helped detect patterns in methylation among different DNA regions

- DMRs were identified using chi-squared test and various criteria during sequencing (mainly pertaining to reading lengths)
- DMR genes were analyzed using GO and KEGG to determine impact of DMR genes on function, and expression, and thus biological ramifications.

#### Correlating DNAM & Gene Expression; Correlating DEGs & DMRs

- Tested given the hypothesis that DNAM helps propagate gene expression, and how the regulation this brings about differs between CO<sub>2</sub> and temp levels.
- Spearman correlation analysis performed. Rho value has to be either positive or negative, p-value below 0.05 to have definitive positive or negative correlation between these two variables.
- Genes were divided based on the level of expression they underwent.
- Methylation rates were quantified separately among the four treatments
- The correlation between methylation of DMRs and gene expression of DEGs was also measured using Spearman correlation analysis and regression analysis.

#### Results:

##### Overall Regulation Among Treatments and Various Functions Impacted

- Among the three experimental treatments, the most overall regulation occurred in the high CO<sub>2</sub> and high temperature treatment, followed by high temperature, then high CO<sub>2</sub> groups
  - ~1100 genes up-regulated and down-regulated in high CO<sub>2</sub> and high temperature
  - ~700-900 genes up-regulated and down-regulated in high temperature
  - ~300-400 genes up-regulated and down-regulated in high CO<sub>2</sub>
- Most genes regulated had to do with catalytic activity, metabolic process, and cells and cell parts.
- Analysis of DEGs reveals that the treatments conditions (in the regulation patterns described above) had heavily regulated metabolic processes.

##### Down-Regulation of Carbon Metabolism Following Long-term High CO<sub>2</sub>

- Enzymes that play a role in glycolysis were significantly down-regulated. Same goes for genes encoding proteins that create the glycolysis pathway.
- Various enzymes and genes relating to pyruvate and its role in respiration were also inhibited under high CO<sub>2</sub> conditions
- Genes producing the enzymes involved in the tricarboxylic acid cycle (TCA) (Krebs Cycle) were also down-regulated.
- Genes involved in the enzymatic methods for concentration of CO<sub>2</sub> waste were likewise inhibited
- Overall, this indicates that the higher CO<sub>2</sub> levels in ocean (and thereby higher pH levels) reduces carbon metabolism due to lowered expression of the genes that produce enzymes and other substances that facilitate the steps of respiration.
- Since carbon metabolism plays a role in amino acid production and metabolism, certain amino acids have been observed to also have been downregulated. Lower amino acid metabolism would reinforce the findings of lower carbon metabolism.

#### Alterations to Amino Acid Metabolism Following Long-term High Temperatures

- As described in figure 2 above, the high temperature treatment experienced the most interference from DEGs, both up and downregulations.
- Comparison between high CO<sub>2</sub> and high temperature treatments: production of 5 amino acids prevented vs. 7
  - Gives examples of how genes involved in producing enzymes that synthesize alanine and histidine are being downregulated.
- However, recall again from figure 2, there was only one upregulated gene for the high-CO<sub>2</sub> treatment. Here, article notes the variations that the high temperature treatment experiences with regards to amino acid metabolism
  - Genes aiding in the synthesis of glycine were upregulated, while for ones metabolizing lysine and cysteine had both up and downregulation

#### Changes to Amino Acid Metabolism Under High Temperatures and High CO<sub>2</sub>

- As for the high CO<sub>2</sub> and high temperature treatments, there is evidence to suggest that these two conditions interact with each other to impact amino acid metabolism
  - One enzyme involved in producing lysine was more upregulated when exposed to high temperature alone than in this treatment, suggesting that the CO<sub>2</sub> inhibited the protein metabolism in this case.
  - Another enzyme that was downregulated with only CO<sub>2</sub> treatment was upregulated in this treatment.
- By contrast, many of the other upregulations and downregulations matched those of high CO<sub>2</sub> and high temperature treatments alone. (there were only four exceptions, and only the ones noted above were discussed)

#### Fatty Acid Metabolism Dynamics following High CO<sub>2</sub>

- Many of the genes producing enzymes involved in fatty acid metabolism were significantly upregulated, which would therefore enhance fatty acid metabolism.
  - This includes a gene producing an enzyme that converts glycerol to glycerol 3-phosphate for making triglycerides, among many others
- However, there were two exceptions (i.e. downregulations)
  - A gene of an enzyme that helps in fatty acid anabolism by establishing double bonds among monounsaturated fatty acids.
  - Another gene which encodes an enzyme (a type of ligase) that helps create cellular lipids
- When carbohydrates cannot be metabolized, lipids and proteins are. Therefore, the increased ease of fatty acid metabolism observed through the overwhelming upregulation of the genes that produce the enzymes involved is an indication of the significant hinderance carbon metabolism faces under these conditions.

#### Down-Regulation of Photosynthesis Following Long-term High Temperatures

- Photosynthetic process, as a whole is inhibited. Genes encoding enzymes and other substances involved in the process of photosynthesis are inhibited.
- Down-regulation of genes that allow for chlorophyll a and b to absorb and use light energy via the creation of light harvesting complexes
- Down-regulation of genes that are involved in producing mechanisms for oxygen discharge during photosynthesis.

- Down-regulation of genes that produces enzymes and other substances that move electrons along the photosynthetic electron transport chain
  - (Unrelated but good to know for photosynthesis) Proton gradient of differing electrical charge is generated as the electrons are transferred to the transport chain. This allows for phosphorylation of ADP via ATP synthase.
- Down-regulation of genes that produces enzyme that helps transfer electrons from the transport chain to help convert NADP into NADPH.
- Production of Mitochondrial ATP Synthase was downregulated while chloroplast ATP synthase remained unaffected.
- Higher temperature seems to be the cause of the reduction in production of ATP synthase, as in high-CO<sub>2</sub> and control treatments, there is no reduction, only ones where temperature is greater.
- Since agents of the light-dependent reaction were inhibited, the Calvin cycle was too (although these two reactions are not always coupled)
- Indeed, most of the genes that produce enzymes and other substances directly involved in Calvin Cycle were inhibited.
- Most genes for the C<sub>4</sub> pathway are also being downregulated, although it is unknown whether *P. tricornutum*
- Overall inhibition of photosynthesis was even observed at the translational stage
- As photosynthetic process was inhibited, the genes producing pigments such as chlorophyll, carotenoids, porphyrins, and aiding in the metabolization of these various pigments were also downregulated.

#### The Impact of Simultaneously High CO<sub>2</sub> and High Temperature More In-Depth

- For the most part, downregulations and upregulations observed were consistent to what was observed for the separate high CO<sub>2</sub> and high temperature treatments.
  - For example, 29 out of 30 genes relating to proteasome, the regulation of the concentration of various proteins and the discarding of folded proteins, were consistently downregulated among all three treatment groups (only 1 was upregulated in the combined treatment)
- However, there were notable differences as well (beyond what has already been noted above):
  - Genes that experienced no impact under high CO<sub>2</sub> or temperature conditions experienced down regulation in the combined treatment, indicating that when these factors act in concert, gene regulation occurs.
  - One gene that was downregulated in the high CO<sub>2</sub> treatment was upregulated in the combined one, suggesting that high temperature prevented inhibition of the gene expression.
  - Conversely, a gene down-regulated in the high temperature treatment was less down-regulated in the combined one, meaning CO<sub>2</sub> helped reduce gene inhibition, with similar patterns being observed for various genes for photosynthesis (although up-regulation was also made less extreme)

#### Transcription and DNAM

- To evaluate the relationship between DNAM and gene transcription, methylation was measured among genomic samples collected.

- While methylation levels vary drastically across the genome, the methylation rates observed matched the well-established data of methylation for the model species.
  - This *should* be the case, however, possibly due to stress of environment and the bisulfite sequencing technique that was used.
- The proportion of various types of methylation, along with genomic rate of methylation was fairly similar among all groups.
  - There were 3 types of DNAM investigated, mCG, mCHG, and mCHH.
  - For mCG, there is a statistically significant negative correlation with gene expression.
  - For mCHG, all but one (assuming  $\alpha = 0.05$ ) have a statistically significant positive correlation with gene expression
  - For mCHH, there are few statistically significant regressions to observe any coherent correlation with gene expression.
- DMRs (standards of determining DMRs enumerated in methodology section) were also investigated.
- Many DEGs occurred in DMRs.
  - Meaning that for the genes were expressed or suppressed to a statistically significant degree, they occurred in regions where there was statistically significant amount of DNA methylation that occurred, indicating that DNA methylation plays major role in gene regulation
  - From least to greatest: High CO<sub>2</sub>, High Temperature, and combined treatments
  - Numerous examples given of various genes that are DEGs that encode proteins for a myriad of processes that are in DMRs.
    - Metabolism of macromolecules, photosynthesis, ribosome biogenesis, among other processes (hey, matches abstract!)

#### Discussion:

- Previous study (done by this group?) showed how there was simultaneously significant genomic diversity losses and enhancements for *P. tricornutum* when exposed to high CO<sub>2</sub> and/or temperature conditions.
- Helped establish understanding of evolutionary biology needed for this study, from there, study is summarized:
  - Goal of showing DNA methylation is empirically connected to gene regulation (novel, as common understanding is DNAM is inhibitory)
  - Predominant finding that most genes were inhibited as a response to these treatment environments. Meaning DNAM causes downregulation of genes!
  - Area of future focus: molecular mechanics of DNAM and changes in gene expression
- Thus, the study argues that DNAM shall serve as an important tool for phytoplankton as they adapt to changing ocean conditions.
- However, some caution:
  - DNAM is not that common and frequent among other phytoplankton species.
  - DNAM might not really regulate gene expression unless in high quantities
  - E.g. larger DNAM was used on same species only to do testing with nitrates.

- Only 18-24% of DEGs occurred in DMR, so DNA methylation may have nothing to do with the gene regulation observed.
- Possibly plays role in regulating sulfate pathways during chromium stress
- Although smaller part of genome for phytoplankton, its possible that DNAM helps metabolism, life cycle, and differentiation of tissue of certain species
- Still, conclusion is that DNAM, even in small rates, can be positively coupled with gene expression.
- Ambiguity persists over whether the traits observed (mainly metabolism) were due to DNA sequence itself changing or epigenetic processes like DNAM.
- Knowing this information is important for tracking biogeography and overall phytoplankton dynamics.
- Via all of those up and downregulations, it was found that phytoplankton can adapt to warmer, more acidic (i.e. water with more CO<sub>2</sub>) conditions by establishing a new optimal metabolic rate, however, this may diminish primary production
- Of all articles so far, this has offered most concrete, realistic advice for helping understand phytoplankton: Long-term studies that examine the evolutionary biological response to stressors ought to occur.
- Establishing links across different levels of biological organization.



## Article #9 Notes: Two-sided effects of the organic phosphorus phytate on a globally important marine coccolithophorid phytoplankton

<b>Source Title</b>	Two-sided effects of the organic phosphorus phytate on a globally important marine coccolithophorid phytoplankton
<b>Source citation (APA Format)</b>	Li, J., Zhang, K., Li, L., Wang, Y., Wang, C., & Lin, S. (2023). Two-sided effects of the organic phosphorus phytate on a globally important marine coccolithophorid phytoplankton. <i>Microbiology Spectrum</i> , 11(5). <a href="https://doi.org/10.1128/spectrum.01255-23">https://doi.org/10.1128/spectrum.01255-23</a>
<b>Original URL</b>	<a href="https://journals.asm.org/doi/full/10.1128/spectrum.01255-23">https://journals.asm.org/doi/full/10.1128/spectrum.01255-23</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	phosphorus nutrient, dissolved organic phosphorus, phytic acid, phytoplankton, coccolithophores, <i>Emiliana huxleyi</i>
<b>#Tags</b>	<a href="#">Fundamental Gene Expression Analysis Process</a> <a href="#">Investigating the Impact of Heavy Metals and Proteins on Phytoplankton and Drawing Connections to Phosphates and Other Compounds</a> <a href="#">Metabolic Ramifications for Higher-Order Organisms</a>
<b>Summary of key points + notes (include methodology)</b>	This study aimed to establish a clearer relationship between varying concentration of phytic acid, a cosmopolitan phosphorous-containing compound and the physiological characteristics of the also cosmopolitan coccolithophore <i>Emiliana huxleyi</i> , which up to this point has not been well understood. Four treatments, including a control, phytic acid, dissolved inorganic phosphorus, and a combined treatment of those two were biochemically analyzed for cell size and concentration, the amount of lipids, photosynthetic output, and nutrient stoichiometry. Gene expression was also analyzed by extracting RNA, converting it into DNA via reverse transcriptional and purification methods, and comparing that DNA to the <i>E. huxleyi</i> to identify DEGs. Findings show that phytic acid upregulated genes relating to carbohydrate, lipid and amino acid metabolism and cell signaling and membrane development, and that, contrary to popular belief in the field, dissolve inorganic phosphorus is not preferentially absorbed over phytic acid. However, an area that still needs further investigation is the potential toxic effects phytic acid may have on phytoplankton as a whole.
<b>Research Question/Problem/ Need</b>	How do varying levels of dissolved organic phosphorous, specifically phytic acid, impact the metabolism and nutrient stoichiometry of phytoplankton?

## Important Figures

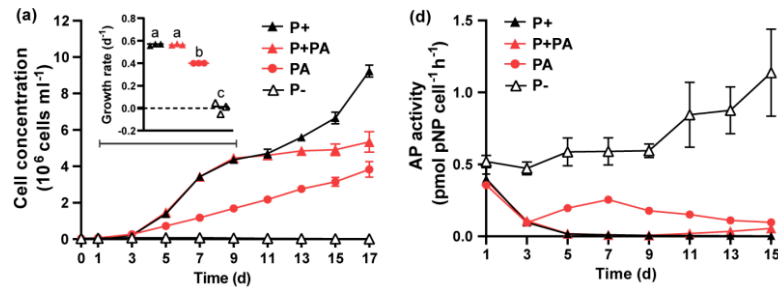
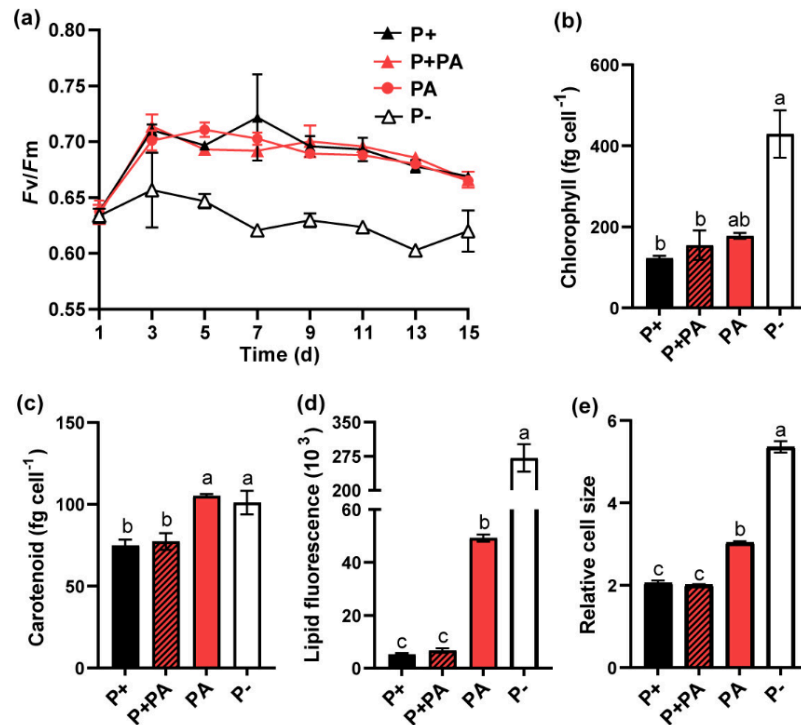


FIG 1 Algal growth and P nutrient of *E. huxleyi* under different P treatments. (a) Growth curves during the experiment and average growth rate (inset) during the exponential growth phase (from days 1 to day 9). (b) DIP concentration in the medium under different P conditions. (c) Cellular particulate P (PP, bar chart) and DIP uptake rate (line chart) on day 1. (d) AP activity. P-, P-depleted; PA, phytate; P+, DIP replete; P+PA, DIP and PA replete. Each data point is the mean of triplicate cultures with the error bar indicating standard deviation (Mean  $\pm$  SD). Samples for transcriptomic analysis were collected on the 5<sup>th</sup> day.

The left line graph shows the cell concentrations among the four groups over the course of the experiment. The cell concentration for the control group remained at a constant 0 cells for 1 million cells per milliliter. Meanwhile, the group exposed solely to DOP (that is, phytic acid, PA), has a linear increase in concentration. Both the groups that were exposed to both DOP and DIP and exclusively DIP followed the same track of concentration, however, at day 11, the former leveled off while the latter grew exponentially.

The right line graph shows the productivity of alkaline phosphatase. In the control environment, where there was no exposure to DOP or DIP, this enzyme became more and more productive (which make sense, as conditions without phosphorous would likely prevent inhibition of a dephosphorylating enzyme). The group exposed to exclusively DIP saw the least productivity, followed by the combined treatment and then the treatment involving exclusively DOP. Conversely to what was observed with cell concentration, it seems that DIP inhibits expression of alkaline phosphatase, whereas DOP allows for it.



**Fig 2** Physiological responses of *E. huxleyi* under different P treatments. (a) Photosynthetic efficiency (Fv/Fm), (b) Chlorophyll content, (c) Carotenoid content, (d) Lipid content measured as fluorescence intensity of BODIPY 505/515 stain. (e) Relative cell size of *E. huxleyi* estimated by normalizing to forward scatter (FSC) of 2  $\mu$ m standard beads. Each data point is the mean of triplicate cultures with the error bar indicating standard deviation (Mean  $\pm$  SD). Different letters above the columns indicate significant differences among groups (ANOVA,  $P < 0.05$ ). Pigments, lipid contents, and relative cell size were measured on the 5<sup>th</sup> day.

Figure 2a illustrates the photosynthetic efficiency of the four treatments over the course of the experiment. Although all of the non-control treatments have slightly declining efficiencies, they are all still above the non-phosphorous containing control. This indicates that while phosphorous boosts photosynthetic activity, the difference in quality between DIP and DOP is negligible.

Although more efficient, figure 2b clearly illustrates that the non-phosphorous treatment had a greater amount of chlorophyll pigments produced than any of the treatments exposed to phosphorous. This is an interesting finding considering that this treatment had the lowest photosynthetic efficiency. Since the control is the least efficient, perhaps less photosynthesis is completed, leading to more chlorophyll pigments being leftover.

Figure 2c, which analyzes carotenoid pigments, might be indicative of a similar pattern. Here, the control and the treatment exposed only to DOPs had the highest carotenoid concentrations. This could indicate that treatments that were exposed to DIP were best able to metabolize, expending both chlorophyll and carotenoids.

Figure 2d compares lipid fluorescence among the four treatments, clearly indicating that the control group exposed to no DOP or DIP had the greatest amount of lipids. While not having as much as the control, the treatment exposed exclusively to DOPs had a much greater amount of lipid fluorescence than the other two groups. The combined treatment has the third highest amount of lipid fluorescence, suggesting that

DIPs inhibit the production of lipids.

Figure 2e, which compares cell size among the four treatments, shows a similar pattern. However, while the control treatment had greater cell sizes on average, here, all three of the experimental treatments are a lot closer when it comes to cell size, although the DOP-exclusive group has considerably larger cell sizes. This once again suggests that DOP might help cell growth, whereas DIP might impede it.

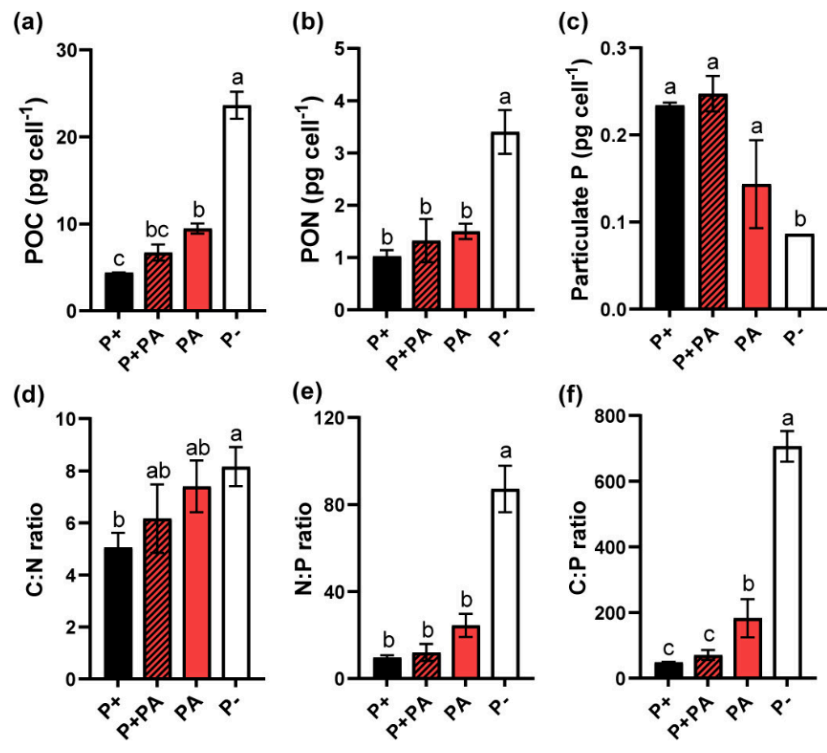
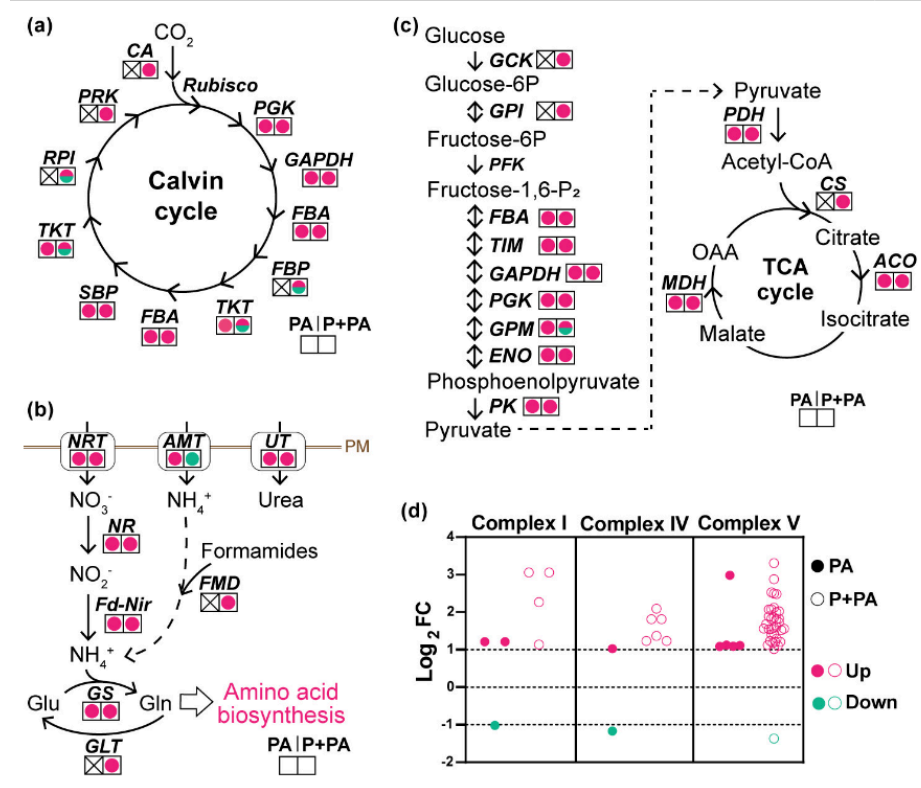


Fig 3 Cellular stoichiometry of *E. huxleyi* under different P conditions measured on the 5<sup>th</sup> day. (a) POC content; (b) Particulate organic nitrogen (PON) content; (c) Particulate P; (d) C:N ratio; (e) N:P ratio; (f) C:P ratio. Each data point is the mean of triplicate cultures with the error bar indicating standard deviation (Mean ± SD). Different letters above the columns indicate significant differences among groups (ANOVA, P < 0.05).

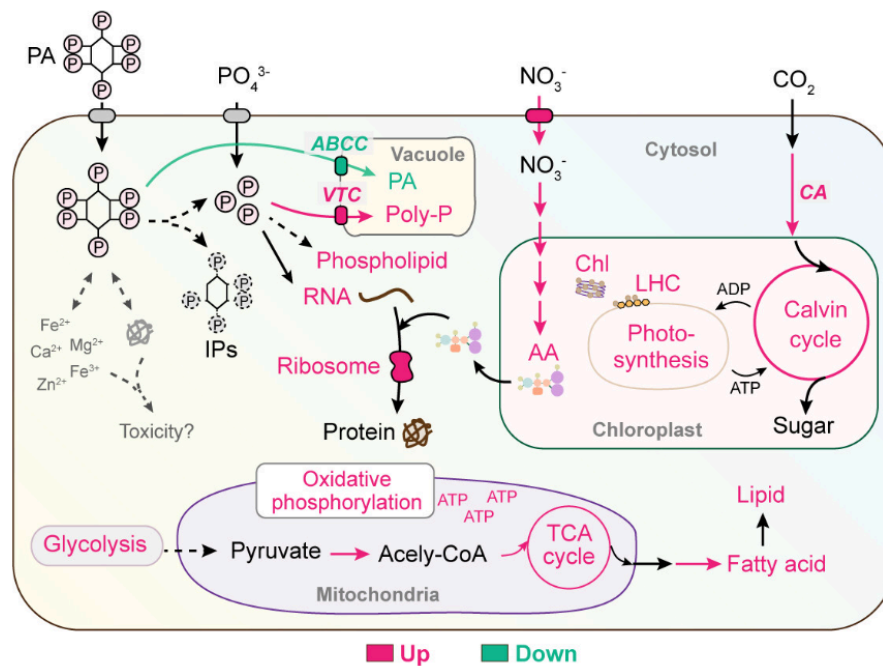
Figures 3a and 3b show an interesting trends regarding particulate organic carbon and nitrogen (POC and PON) levels. In both cases, the control has a significantly greater amount of both particulates. This means that carbon and nitrogen absorption occurred to the least extent for the control group, implying that exposure to phosphorous assists phytoplankton in incorporating these compounds. However, following this logic, since the POC and PON levels are higher in the POC-exclusive treatment, followed by the combined treatment and then the PIC-exclusive treatment, this suggests that PICs maximize carbon and nitrogen integration.

Based of the scaling of each graph figures 3d through 3f, the ratio between carbon, nitrogen, and phosphorus is such that carbon is the most plentiful, followed by nitrogen, and then phosphorus. Otherwise, seeing as the control has little phosphorus to begin with, it make sense that it has the C:P and N:P highest ratios. However, from C:N, N:P and C:P, the ratios became more varied, likely due to the abundance levels of each substance.



**Fig 4** Transcriptomic responses of *E. huxleyi* to PA utilization. (a) DEGs related to Calvin cycle in PA/P+ and (P+PA)/P+ comparisons. CA, carbonic anhydrase; Rubisco, rubisco ribulose-1,5-bisphosphate carboxylase; PGK, phosphoglycerate kinase; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; FBA, fructose-bisphosphate aldolase; FBP, fructose-1,6-bisphosphatase; TKT, transketolase; SBP, sedoheptulose-bisphosphatase; RPI, ribose 5-phosphate isomerase; PRK, phosphoribulokinase. (b) DEGs related to nitrogen uptake and assimilation in PA-grown *E. huxleyi* cells relative to cells in the P+ group. PM, plasma membrane; NRT, nitrate transporter; AMT, ammonium transporter; UT, urea transporter; NR, nitrate reductase; Fd-Nir, ferredoxin-nitrite reductase; FMD, formamidase; GS, glutamine synthetase; GLT, glutamate synthase. (c) DEGs related to EMP-tricarboxylic acid cycle (TCA) pathway in PA-containing groups compared to the P+ group. GCK, glucokinase; GPI, glucose-6-phosphate isomerase; PFK, phosphofructokinase; FBA, fructose-bisphosphate aldolase; TIM, triosephosphate isomerase; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; PGK, phosphoglycerate kinase; GPM, phosphoglycerate mutase; ENO, enolase; PK, pyruvate kinase; PDH, pyruvate dehydrogenase; CS, citrate synthase; ACO, aconitate hydratase; MDH, malate dehydrogenase; OAA, oxaloacetate. (d) Regulation of oxidative phosphorylation in PA and P+PA groups relative to the P+ group. Complex I includes NADH dehydrogenase, Complex IV includes cytochrome c oxidase, and Complex V includes F-type ATPase, V-type ATPase, H<sup>+</sup>-transporting ATPase, and inorganic pyrophosphatase. The left and right squares indicate expression regulation in PA/P+ and (P+PA)/P+ comparisons, respectively. Berry and lawngreen cycles, respectively, indicate significant upregulated and downregulated in PA/P+ and (P+PA)/P+ comparisons. Crossed squares indicate no significant differential expression in PA/P+ or (P+PA)/P+ comparisons.

This figure illustrates the various upregulations and downregulations of different metabolic processes. Red indicates up, green down. Given the overwhelming presence of red circles, it is clear that most genes are upregulated. As a result, this indicates that PA exposure leads to more efficient metabolism.



**Fig 5** Proposed PA utilization mechanism in *E. huxleyi*. Berry and lawngreen words (and arrows) indicate upregulated and downregulated processes, respectively, in PA-grown *E. huxleyi* cells. PA, phytate; IPIs, inositol phosphates; Poly-P, polyphosphate; LHC, light-harvesting complex; AA, amino acid; VTC, vacuolar transport chaperone; ABCC, ATP-binding cassette subfamily C; CA, carbonic anhydrase. The encircled P represents phosphate.

This is another diagram that illustrates (more precisely) the predicted changes in metabolic processes, with red indicating upregulation and green downregulation. Given the locations of the red text on this diagram, it is clear that carbohydrate metabolism (both photosynthesis and respiration) are to be enhanced due to significant upregulation. The same can be said about amino and fatty acid metabolism, as well as ribosomal and RNA production and phospholipid interactions.

#### VOCAB: (w/definition)

**Phosphoester** - A phosphorus-containing molecule bounded to two hydroxyl groups, two oxygens, one being a double bond), and the other bonded to a replacement group. These are converted into phosphodiester by replacing the hydroxyl with another oxygen bonded to a replacement group. This molecule forms bonds that connect nucleotides, forming DNA/RNA.

**Phosphonate** - A salt-like phosphorus containing molecule where P is bonded to carbon, oxygen, hydrogen and hydroxyl groups.

**Alkaline phosphatase** - An enzyme responsible for dephosphorylating compounds.

**mRNA enrichment** - A process through which mRNA can be isolated. This involves attaching a bead to a poly-A tail of mRNA, and then detaching the rest of the mRNA from this tail-bead bond. This allows for more precise focus on less contaminated RNA molecules.

**Reverse Transcription** - The production of DNA from mRNA.

#### Cited references to follow up on

Shemi A, Schatz D, Fredricks HF, Van Mooy BAS, Porat Z, Vardi A. 2016. Phosphorus starvation induces membrane remodeling and recycling in *Emiliania*

	<p>huxleyi. <i>New Phytol</i> 211:886–898.</p> <p>Benner I, Passow U. 2010. Utilization of organic nutrients by coccolithophores. <i>Mar Ecol Prog Ser</i> 404:21–29.</p> <p>Shaked Y, Xu Y, Leblanc K, Morel FMM. 2006. Zinc availability and alkaline phosphatase activity in <i>Emiliana Huxleyi</i>: implications for Zn-P Co-limitation in the ocean. <i>Limnol Oceanogr</i> 51:299–309.</p> <p>Li J, Zhang K, Lin X, Li L, Lin S. 2022. Phytate as a phosphorus nutrient with impacts on iron stress-related gene expression for phytoplankton: insights from the diatom <i>Phaeodactylum tricornutum</i>. <i>Appl Environ Microbiol</i> 88:e0209721.</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How do various molecular micronutrients interact with each other to influence phytoplankton metabolism and cellular physiology?</li> <li>2. How do the varying cytological attributes of different phytoplankton species influence the optimum phosphate levels under which they can operate?</li> <li>3. How do varying levels of phosphate and phosphorus-containing compounds impact the rate of cellular growth and the cell cycle? What implications does this have for metabolism?</li> <li>4. Does an enhancement in metabolic processes among phytoplankton result in an enhancement in metabolic processes among other organisms higher up the trophic pyramid?</li> <li>5. What biochemical interactions does phytic acid undergo to cause the upregulation of genes that end up enhancing metabolism and other physiological functions?</li> </ol>

## NOTES

### Abstract:

- Phosphorous is a potential agent of eutrophication
- Inositol hexaphosphate, aka phytic acid (hereafter PA) is a major oceanic nutrient, but mechanisms and impact on phytoplankton not well understood
- PA, when the only source of P, plays a major role in enhancing metabolic processes, C & N fixation, and amount of lipids.
  - P also plays major role in various parts of homeostasis and community structure
- However, PA can also be toxic
- These findings are important in understanding the relationship between P and phytoplankton conditions
- The use of common DOP (dissolved organic phosphorous compound) was analyzed on a common species of phytoplankton.
- Both physiological and transcriptomic means of measurement and analysis.

### Introduction:

- Mentions difference between DOP and DIP (dissolved inorganic phosphorus compound), risk of P causing eutrophication. DOP > DIP in concentration

- Two types of DOP: phosphoester (80% of DOP), and phosphonate (20% of it)
- PA, which is a phosphoester, cannot easily be absorbed by many land plants due to lack of enzymatic capabilities. This leads to run-off into the ocean as this is excreted as aqueous waste, and thus, risks for eutrophication
- Eutrophication leads to different, disrupted community structure.
- *Emiliana huxleyi* (*E. huxleyi*) is the most cosmopolitan and abundant coccolithophore
- Past work has shown that other phosphorus-containing molecules have been successful for this species, but PA and other compounds and species need to be investigated.
- PA is a good mechanism for growth and helps combat starvation of P
- Findings show large amounts of change in metabolic activity in order to adapt to varying P levels.

#### Means and Methods:

- Incubation: 14 hour light, 10 hours dark cycle in seawater.
- During the exponential growth phase of the algae (*E. huxleyi*), were extracted, deprived of all P, and the exposed to four following conditions in a media
  - No dissolved inorganic or organic phosphorus (sort-of a control) (**P-**)
  - 36  $\mu$ M of Dissolved Inorganic Phosphorus (**P+**)
  - 12  $\mu$ M Phytic Acid (the DOP used in the experiment) (**PA**)
  - 12  $\mu$ M Phytic Acid and 12  $\mu$ M Dissolved Inorganic Phosphorus (**P+PA**)
- Use of microscopes to track growth and cell size, along with flow cytometer.
  - Calculations used to track growth rate (original and final population,  $\ln()$ )
- In order to measure photosynthetic output, all four groups had sample of 10m cells taken, put into a dark solution of methanol. Then, chlorophyll and carotenoid pigments were extracted and measured using spectrophotometer.
- DIP concentrations kept constant to the levels assigned through heating mechanisms and potassium persulfate.
- Alkaline phosphatase (see definition) activity was measured by taking samples, exposing them to 2 hours of darkness, followed by centrifuging process. Using nanowaves, absorbance rates were measured.
- In order to measure carbon fixation and nitrogen assimilation, all samples were cooked in oven, fumed in HCl, and placed in tinfoil sheets. From there, an element analyzer machine was used.
- Amount of Lipid quantified by taking sample of 1m cells, staining it, leaving it in the dark for 20 min, and then using flow cytometer, measuring fluorescence
- For analyzing gene expression: 50m cells from each treatment were centrifuged. Then, RNA was extracted, subject to enrichment (see definition above), reverse transcription, the re-addition of the poly-A tails, Okazaki fragments were ligated, then PCR amplification, denaturation and cyclization were used to create a DNA sequence.
- Low quality nucleotides were eliminated, and the remaining ones were compared to the *E. huxleyi* genome to measure gene expression.
- Using KEGG and DEG analyses and a false discovery rate of  $q < 0.05$ , differentially expressed genes were identified
- ANOVA, means, stdev, were used for statistical analyses.
- 17 day experiment

Results (see figures section, they're basically synonymous, given the article's structure):



- The cell concentration for the control group remained at a constant 0 cells for 1 million cells per milliliter. Meanwhile, the group exposed solely to DOP (that is, phytic acid, PA), has a linear increase in concentration. Both the groups that were exposed to both DOP and DIP and exclusively DIP followed the same track of concentration, however, at day 11, the former leveled off while the latter grew exponentially.
  - Barring the control (which I think is meant to be at 0 concentration), this suggests that DIP help increase cell growth and concentration, whereas DOP decreases it. This would explain why the concentration eventually leveled off for the combined treatment
- For Alkaline Phosphatase:
  - In the control environment, where there was no exposure to DOP or DIP, this enzyme became more and more productive (which make sense, as conditions without phosphorous would likely prevent inhibition of a dephosphorylating enzyme). The group exposed to exclusively DIP saw the least productivity, followed by the combined treatment and then the treatment involving exclusively DOP. Conversely to what was observed with cell concentration, it seems that DIP inhibits expression of alkaline phosphatase, whereas DOP allows for it.
- For photosynthetic efficiency of the four treatments over the course of the experiment, although all of the non-control treatment have slightly declining efficiencies, they are all still above the non-phosphorous containing control. This indicates that while phosphorous boosts photosynthetic activity, the difference in quality between DIP and DOP is negligible.
- Although more efficient, the non-phosphorous treatment had a greater amount of chlorophyll pigments produced than any of the treatments exposed to phosphorous. This is an interesting finding considering that this treatment had to lowest photosynthetic efficiency. Since the control is the least efficient, perhaps, since less photosynthesis is completed, more chlorophyll pigments are leftover.
- Data on the caretenoid pigments might be indicative of a similar pattern. Here, the control and the treatment exposed only to DOPs had the highest caretenoid concentrations. This could indicate that treatments that were exposed to DIP were best able to metabolize, expending both chlorophyll and caretenoids.
- As for lipid fluorescence among the four treatments, it is clear that the control group exposed had the greatest amount of lipids. While not having as much as the control, the treatment exposed exclusively to DOPs had a much greater amount of lipid fluorescence than the other two groups. The combined treatment has the third highest amount of lipid fluorescence, suggesting that DIPs inhibit the production of lipids.
- Figure 2e, which compares cell size among the four treatments, shows a similar pattern. However, while the control treatment had greater cell sizes on average, here, all three of the experimental treatments are a lot closer when it comes to cell size, although the DOP-exclusive group has considerably larger cell sizes. This once again suggests that DOP might help cell growth, whereas DIP might impede it.
- With regards to particulate organic carbon and nitrogen (POC and PON) levels, in both cases, the control has a significantly greater amount of both particulates. This means that carbon and nitrogen absorption occurred to the least extent for the control group, implying that exposure to phosphorous assists phytoplankton in incorporating these

compounds. However, following this logic, since the POC and PON levels are higher in the POC-exclusive treatment, followed by the combined treatment and then the PIC-exclusive treatment, this suggests that PICs maximize carbon and nitrogen integration.

- The ratio between carbon, nitrogen, and phosphorus is such that carbon is the most plentiful, followed by nitrogen, and then phosphorus. Otherwise, seeing as the control has little phosphorus to begin with, it makes sense that it has the C:P and N:P highest ratios. However, from C:N, N:P and C:P, the ratios became more varied, likely due to the abundance levels of each substance.
- For the transcriptional analysis, of the 36,293 genes that were identified as expressed, 4,153 were considered DEGs, specifically when comparing the DOP-exclusive treatments with the DIP-exclusive treatment. This corresponded to 3 KEGG metabolic pathways related to amino acid metabolism and carbon fixation.
- However, when comparing the combined treatment to the DIP-exclusive treatment, there were 6,412 DEGs, mapped to 19 metabolic pathways, which, in addition to the ones above, included oxidative phosphorylation, photosynthesis, nitrogen and fatty acid metabolism, and so much more.
- Genes associated with carbon fixation and photosynthesis were significantly upregulated. This included genes related to electron transport chain, ATP synthase, light-harvesting complex, as well as numerous genes related to the Calvin cycle. Carbonic anhydrase, the main mechanism of carbon fixation, is upregulated.
- For amino acid metabolism, increased production and abundance of ribosomes, along with the upregulation of various enzymes responsible for the catabolizing and anabolizing various nitrogenous and other protein compounds was observed.
- Meanwhile, for carbohydrate metabolism, genes involved in Krebs cycle and glycolysis were also significantly upregulated.
  - These various upregulations were observed primarily in PA (DOP)-exclusive and combined treatments.

#### Discussion:

- Reiterates purposes of study, to understand specifically the impact of phytic acid (PA). References cosmopolitan scope of both the compound and *E. huxleyi*.
  - Study with a previous diatom showed that phytic acid impeded iron availability.
  - Argues that *E. huxleyi* are versatile in adapting to different conditions, including the different DIP and DOP levels of this experiment.
  - PA use could play a major role in nutrient cycling done by phytoplankton
- In groups that contained the DOP-exclusive (PA) treatment, there was no detection of DIPs, indicating that PA could be easily absorbed *E. huxleyi* by cells.
- However, observation of the DIP-containing treatments indicated that DIP had no impact of the ability for PA to be absorbed, which is surprising, b/c expectation is that DIP would be absorbed in favor of PA.
- Once absorbed, the PA serves as an important source of growth.
- Study investigates whether PA can be absorbed by *E. huxleyi* (it can indeed be absorbed by terrestrial plants), comparing it to *Phaeodactylum tricornerutum*. Given the down regulation of certain genes, it is concluded the intracellular hydrolysis and utilization are

the main sinks for *E. huxleyi*, whereas for *P. tricornutum*, PA causes less growth due to greater extracellular hydrolysis and vacuolar storage.

- Different ways of using nutrients like PA have important ecological ramifications regarding phenology, bloom patterns, and succession that are caused by changing phosphorous levels.
- As we've seen, metabolism is impacted by the use of PA (the DOP). RNA is in fact an indicator of P nutrient quota and status, so the increased RNA points to how PA is being increasingly incorporated by *E. huxleyi*
- PA is used to enhance synthesis of Poly-P, an important molecule involved in cell signaling. In a similar vein, genes for the metabolism of phospholids were also upregulated, meaning PA plays an important role in strengthened cell membranes, signaling, and communication.
- Additionally, the PA-grown treatments caused greater amount of phosphorylation, enhancing the production of ATP through facilitating production of ATP synthase.
- These trends represent intriguing allocation of PA on part of *E. huxleyi*
- The C:P, N:P, and C:N ratios have all increased due to improved ability to fix carbon and assimilate nitrogen. The higher ratios underscore the enhancements to the metabolic processes for phytoplankton.
- To test for PA-induced toxicity (something observed in their previous study w/ *P. tricornutum*), stressors between the exclusively-PA and combined treatments were compared.
- Both PA-containing groups displayed same signs of metabolic stress, namely greater amounts of lipids and enhanced fatty acid metabolism. Genetic adaptations also included faster rates of metabolism, which is also associated with coping with environmental strains. Given that these stressors are the same despite the presence of DIP, this indicates that PA can potentially be toxic.
- PA is known for its attraction to various heavy metals (Fe, Mg, Ca, Zn), and restricting access to essential minerals and proteins.
- The lower growth rates observed in the PA-exclusive treatment could indicate toxicity
- More research needs to be done on heavy metals and proteins in relation to PA on phytoplankton.

#### Conclusion:

- PA is used to support growth (although it may be toxic and slow down growth in the process. This needs more research)
- DIP is not preferentially absorbed over PA by *E. huxleyi*, which challenges a long-held belief that they were. Moreover, DIP (which to clarify, was not in itself a variable, rather it was used in treatments to test the main variable, which was PA), does not impact the absorption of PA.
- Reiterates enhanced metabolic processes and other key findings

## Article #10: Three-dimensional model for analysis of spatial and temporal patterns of phytoplankton in Tucuruí reservoir, Pará, Brazil

<b>Source Title</b>	Three-dimensional model for analysis of spatial and temporal patterns of phytoplankton in Tucuruí reservoir, Pará, Brazil
<b>Source citation (APA Format)</b>	Deus, R., Brito, D., Kenov, I. A., Lima, M., Costa, V., Medeiros, A., Neves, R., & Alves, C. N. (2013). Three-dimensional model for analysis of spatial and temporal patterns of phytoplankton in Tucuruí Reservoir, Pará, Brazil. <i>Ecological Modelling</i> , 253, 28–43. <a href="https://doi.org/10.1016/j.ecolmodel.2012.10.013">https://doi.org/10.1016/j.ecolmodel.2012.10.013</a>
<b>Original URL</b>	<a href="https://www.sciencedirect.com/science/article/abs/pii/S0304380012005157">https://www.sciencedirect.com/science/article/abs/pii/S0304380012005157</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Modeling, Reservoir eutrophication, MOHID (abbreviation of Portuguese for “Hydrodynamic Model”), Water quality, Management
<b>#Tags</b>	<a href="#">Comparing Results of Different Computer Modeling Techniques and Identifying Methods that Optimize Prediction Accuracy</a> <a href="#">Various Parameters In Modeling Phytoplankton Populations</a>
<b>Summary of key points + notes (include methodology)</b>	This article aimed to develop a computer model that delineates spatial and temporal patterns in phytoplankton populations and correlate those findings to biological factors including DO, micronutrients, light availability, chlorophyll a concentrations, as well as hydrodynamical and meteorological factors for the Tucuruí reservoir in Brazil. A model known as the MOHID, comprised of a hydrodynamic module based off the Navier-Stokes equation and a water quality module based off multiple parameters relating to biological and meteorological conditions, was compared to field data collected from 2010-2011, which included temperatures at varying positions along the vertical profile of the reservoir, and biological conditions at two stations, both at the surface and bottom levels. The model was validated, given that, when comparing predictions to the field data, NOF values were near 0, and $R^2$ and gamma values were near 1. Although the model found that the reservoir was phosphorus-limited, and, given the concentrations of DO, micronutrients, and chlorophyll, mesotrophic, the hydrodynamics of the reservoir were the limiting factor in phytoplankton growth. Namely, the rapid inflow and outflow of the reservoir created rapid residence times which inhibited phytoplankton growth.
<b>Research Question/Problem/Need</b>	How can a computer model that uses the hydrodynamical, meteorological, and biological factors of the Tucuruí reservoir be developed and validated in order to elucidate spatial and temporal trends in phytoplankton and establish correlations between these factors and phytoplankton populations?

## Important Figures

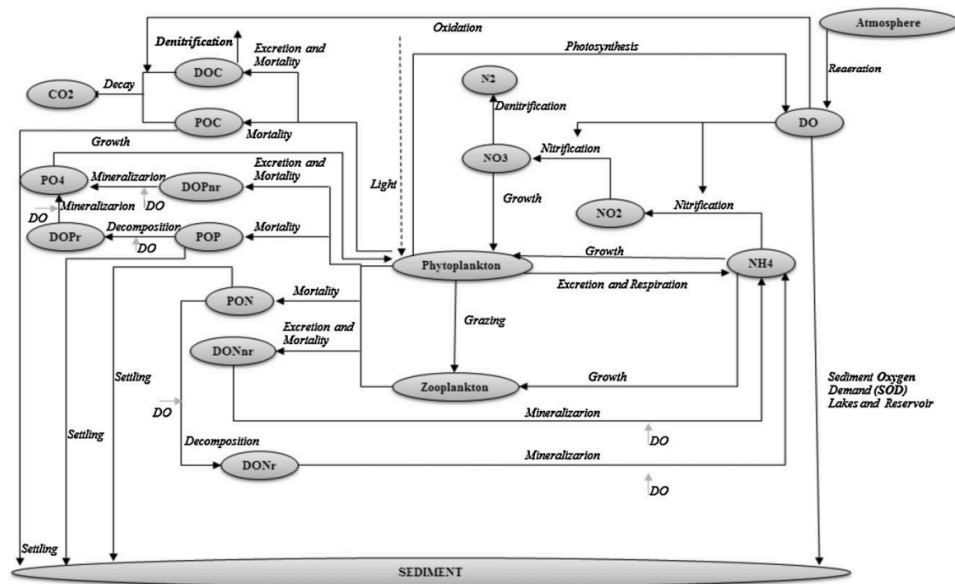
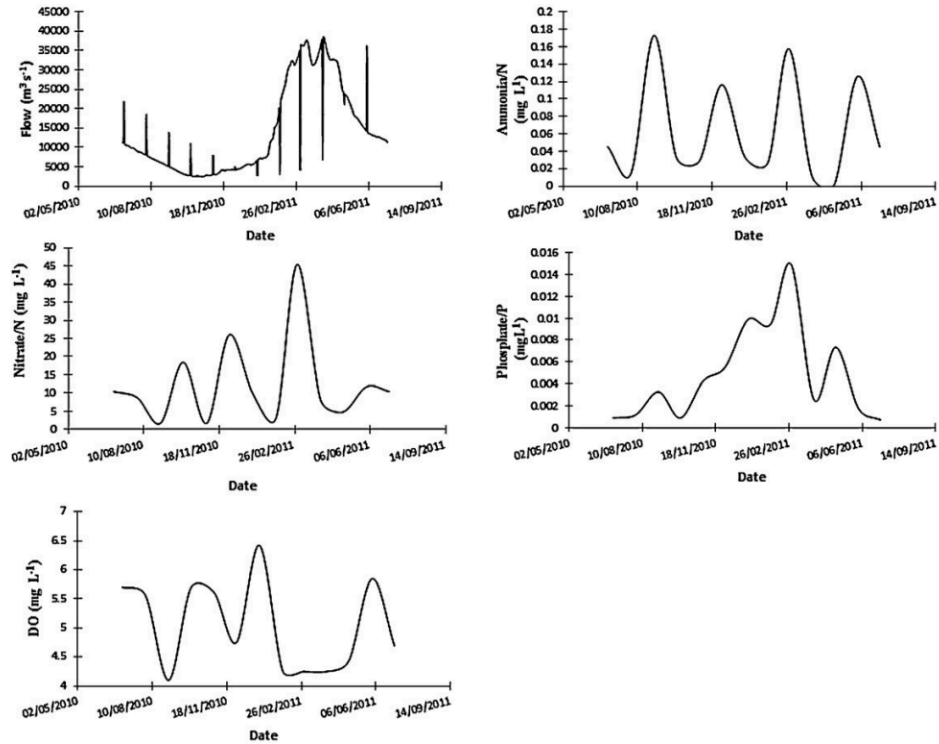


Fig. 2. Conceptual model of MOHID water quality module.

This diagram comprehensively illustrates biogeochemical cycling in a water body. Starting from the atmosphere, oxygen is absorbed via reaeration. Notice that the oxygen is DO, which, through various processes, is implemented into respective places in the cycle through the respective processes. Some DO becomes sedimentation, due to Sediment Oxygen Demand (SOD). Looking at another critical center point in the diagram, light allows for phytoplankton carry out photosynthesis. Through excretory processes and respiration, NH<sub>4</sub> is released. Nitrification occurs as NH<sub>4</sub> is converted into NO<sub>2</sub>, and NO<sub>2</sub> into NO<sub>3</sub>. Through these processes, DO is implemented. Then, NO<sub>3</sub> is denitrified into N<sub>2</sub>. NH<sub>4</sub> also contributes to the growth in the phytoplankton. Via grazing, nutrients are transferred to zooplankton (which is also able to grow via NH<sub>4</sub>). Both zooplankton and phytoplankton, through both excretion and mortality, produced dissolved organic nitrogen and dissolved organic phosphorus. However, the formation of particulate organic nitrogen and phosphorus (particulate meaning concentrated into one entity, opposite of dissolved) occurs exclusively through mortality. Additionally, particulate organic carbon only forms from phytoplankton mortality, and only through mortality or excretion of phytoplankton is dissolved organic carbon formed. Particulate organic phosphorus, nitrogen and carbon all, through settling process, sink and contribute to sedimentation. Particulate organic nitrogen decomposes into dissolved organic nitrogen via the incorporation of DO (which occurs for processes for phosphorus and nitrogen). DON is then used to form NH<sub>4</sub> via mineralization. Similarly, particulate organic phosphorus becomes dissolved through decomposition, and all DOP is mineralized into PO<sub>4</sub>, which, like NH<sub>4</sub>, serves as nutrient of growth for phytoplankton. Lastly, through decay, all POC and DOC is converted into CO<sub>2</sub> and released into the atmosphere.

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These figures illustrate the raw, field data collected 2010-2011 (which was then to be compared to the computer models) for flow rate, ammonium, nitrates, phosphates, and DO. The flow rate appears to be faster between February and August, and slower between August and February. The slowest rate appears to be around October 2010, with a rate of around  $3000 \text{ m}^3/\text{s}$ . Meanwhile, all the nutrients oscillate at much faster rates and with much more unique patterns. For ammonium, the oscillation patterns seems to be that it reaches minimum concentrations between about 0.00 and 0.03  $\text{mg/L}$ , reach one high of between about 0.10-0.13  $\text{mg/L}$ , and another at around 0.16  $\text{mg/L}$ , with these two peaks alternating between each other. For nitrates, there is also an oscillatory pattern, however, each peak is greater, reaching about 15, 25 and then 45  $\text{mg/L}$ . Phosphate concentration starts off at low concentrations, around 0.002 to 0.004  $\text{mg/L}$ . Then, between November and February, it increases gradually to about 0.015  $\text{mg/L}$ , before declining (with another upward jump) back down to the lower levels. Lastly, DO tends to hover between 5.5 and 6.5  $\text{mg/L}$ , with the exception of two oscillations downward to about 4.1  $\text{mg/L}$ , and another two to about 4.7 to 5.0  $\text{mg/L}$ .

This shows that each nutrient (in conjunction with flow rate), fluctuates at different rates, and that their concentrations vary greatly.

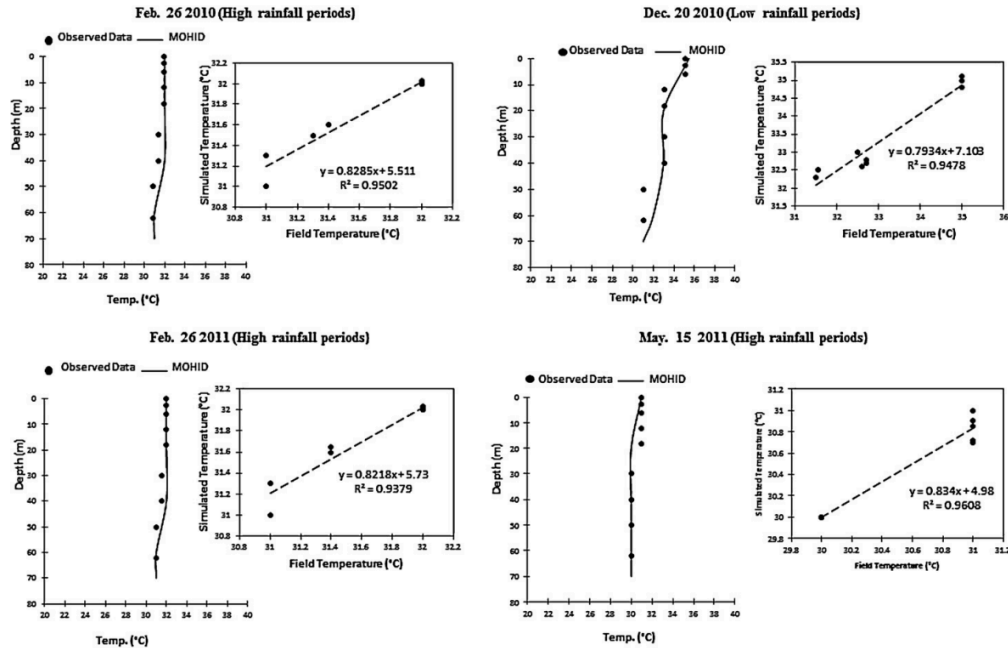


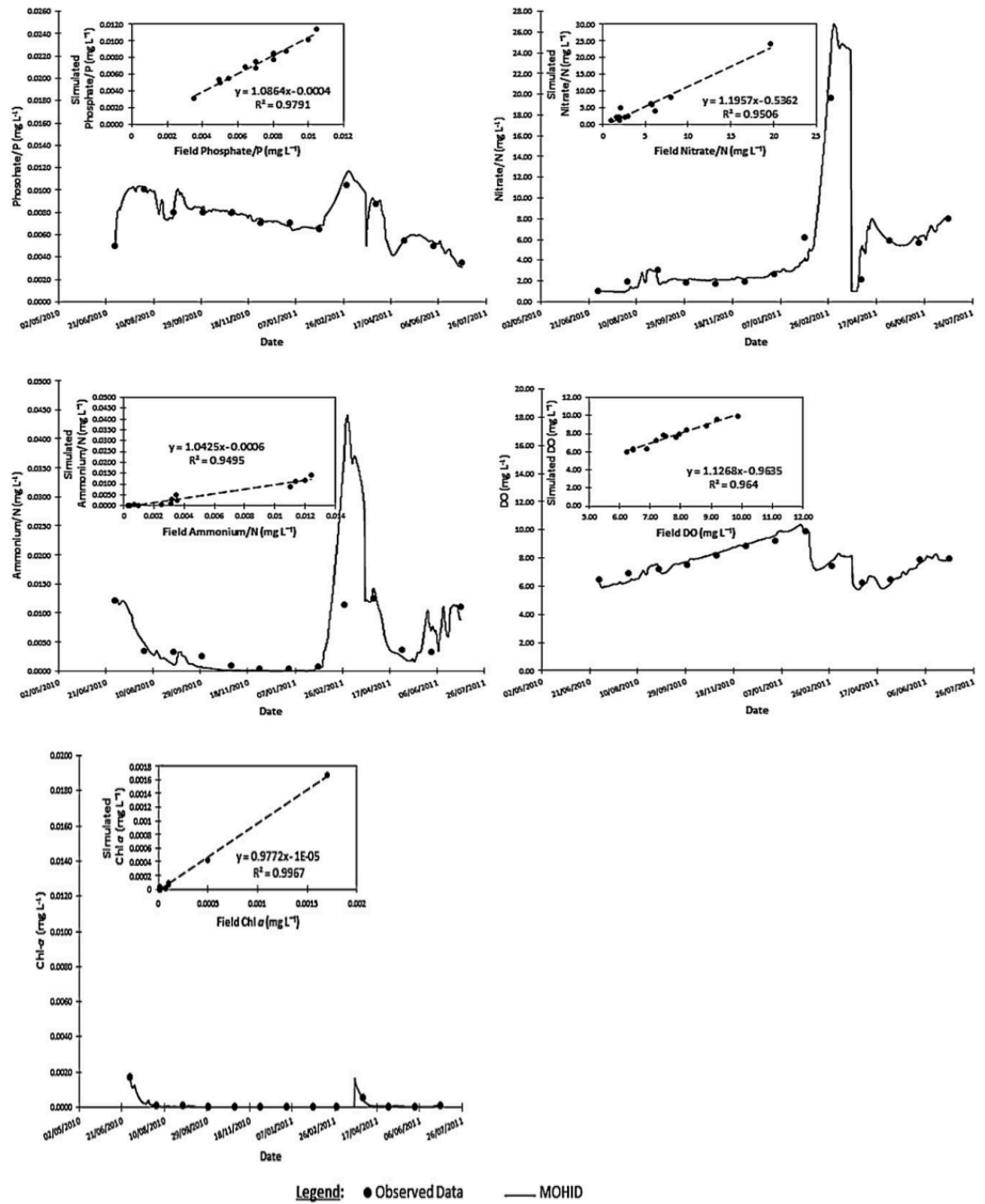
Fig. 7. Simulated vertical profiles of temperature vs. field data at M1 station, Tucuruí reservoir, during low and high rainfall period.

Table 2

Statistical applied to simulated vertical profiles of temperature vs. field data at M1 station, Tucuruí reservoir, during low and high rainfall period.

Monit point	Parameter	February 26, 2010 (high rainfall periods)	December 20, 2010 (low rainfall periods)	February 26 2011 (high rainfall periods)	May 15 2011 (high rainfall periods)
	NOF	0.0080	0.0226	0.0095	0.0091

The various scatter graphs and accompanying table below display the comparisons of the model predictions and field data of temperature at the different vertical profiles. From these data and figures, it is very clear that the model has pretty accurate predictions. For all four instances of measurement (wet seasons and dry seasons 2010 & 2011), when comparing the raw data to the predicted values (i.e. the left graph in each quadrant), visually, the data plots of the actual field data do not stray that far away from the lines representing the predicted temperatures from the model. More quantitatively, looking at the scatterplots at each for each of the four instances (the ones on the right in each quadrant), we can observe more evidence of proper model calibration/good accuracy for predicting these trends. The gamma, i.e. the slopes, chronologically (high, then low rainfall 2010, and again for 2011), were 0.8285, 0.7934, 0.8218 and 0.834, meaning, overall, the model underestimated temperature at varying points along the vertical profile by about 20%. However, note that there are only a few data points. Thus, gamma (and thereby R<sup>2</sup>, and too an extent, entire linear model and NOF) is a weak indication of model efficacy. Gamma is the most “vulnerable” statistic to small sample size (because leverage points/outliers are going to be more drastic), other variables impacted too, but not as much. Either way, all R<sup>2</sup> values are above 0.93. These high R<sup>2</sup> metrics indicate that the variation in the predicted values are strongly based off the actual field data. Similarly, all NOF values are within 0.0300 of 0.0000, meaning the average of the difference between each individual predicted value from each field data value was nearly 0. Overall, there is considerable hegemony between the model predictions and the field data, indicating that in this respect, the model produces accurate results.



This series of graphs, in conjunction with the table below, from top left, display the comparisons of model prediction and field data for phosphates, nitrates, ammonium, DO, and chlorophyll a concentrations, all in mg/L, for the bottom of the reservoir at one of two stations, specifically M3 (therefore four total sets of graphs like these that display similar result as to what is about to be discussed). The values for R<sup>2</sup>, gamma slope, and NOF for all five parameters indicate that the model is rather accurate. Qualitatively, with the exception of a few outliers (for example, ammonium levels in Early March 2011 were severely overestimated, similar thing with phosphates and nitrates), all data points are very



close, if not, visually on the line representing computer predictions. Quantitatively, going from the top left, the gamma slopes are 1.0864, 1.1957, 1.0425, 1.1268, and 0.9772. This suggests that, for predictions of this model in this area, nitrate and DO levels are most heavily overestimated for nitrates and ammonium (I don't know if it's ammonia or ammonium we're looking at in this study). However, among the four areas tested, while the slopes indicated the the model did overestimate a little bit, consider that small sample size, combined with outliers and leverage points created and accentuated these abnormalities. Of the four (station 1/3, surface/beneath), this one had the worst prediction rate when looking exclusively at slope. By contrast, the  $R^2$  and NOF values were most ideal for the four tests of the model. From the top left, they are 0.9791, 0.9506, 0.9495, 0.964, and 0.9967. These high values suggest that all the computer modeling predictions are based almost entirely off (i.e. agrees with and properly simulates) the field conditions. It also demonstrates that some of these links are stronger for some nutrients and factors than others (and this itself would vary site to site). Lastly, from left to right, 0.0563, 0.3504, 0.2398, 0.0349 and 0.1667 represent the NOF for these parameters. All of these values are somewhat close to 0.0000, and although some are of a considerable numerical difference, all values are between 0.0000 and 0.10000 are considered signs that the data predicted from the model is in good match with the actual field data. Low NOF values indicate the difference between individually predicted and actual values, or at least the variation in said quantity (RMSE), is very little. Taken together, these data and findings clearly show that the model created is worthy of validation (which it did get).

**Table 3**

Statistical applied to verification model calculated phosphate, nitrate, ammonia, dissolved oxygen and chlorophyll *a* concentrations vs. surface and bottom field data in the Tukurui reservoir (M1 and M3 Stations).

Monit. points	Parameter	Statistical verification				
		Phosphate/P	Nitrate/N	Ammonia/N	OD	Ct/a
M1 Station surface	NOF	0.2983	0.1901	0.4453	0.0792	0.4494
	$\gamma$	0.9688	0.9493	1.2172	1.1319	1.0540
	$R^2$	0.9525	0.9448	0.9235	0.9240	0.9388
M1 Station bottom	NOF	0.1389	0.2135	0.2792	0.1173	0.4466
	$\gamma$	0.9027	0.9149	1.1535	1.0833	1.1872
	$R^2$	0.9668	0.9782	0.9570	0.9665	0.9085
M3 Station surface	NOF	0.1034	0.1247	0.1376	0.0627	0.4932
	$\gamma$	0.9259	1.0060	0.9556	0.7627	0.9975
	$R^2$	0.8921	0.9791	0.9786	0.7946	0.8993
M3 Station bottom	NOF	0.0563	0.3504	0.2398	0.0349	0.1667
	$\gamma$	1.0864	1.1957	1.0425	1.1268	0.9772
	$R^2$	0.9791	0.9506	0.9495	0.9640	0.9967

Lastly, this table [...]

**VOCAB:  
(w/definition)**

**Process-oriented Modeling** - A technique that represents one or more variables, and how they are impacted by one or more processes as a means of characterizing an entire system and its dynamics. This technique operates on an input-output basis, and involves partial differential equations, the implementation of empirical data, and other advanced tools.

**Advection** - The movement of nutrients and other material via fluid currents.

**Water Mean Residence Time** - In the context of this study, this is equal to the total amount of water in the reservoir divided by either the inflow or outflow. More broadly, it is the measure of time a portion of a fluid stays in some controlled container.

**Navier–Stokes Equations** - A set of four partial differential equations that model the

movement of non-compressible fluids.

**Water Column Light Extinction Coefficient** - The proportion of surface water light that is absorbed and scattered within a one meter long vertical depth in the water.

**Reaeration** - The absorption of atmospheric material into a body of water. In the context of this study, this refers to the absorption of O<sub>2</sub> in order to replenish DO, a process which may be carried out via artificial means.

**Sediment Oxygen Demand** - The rate at which DO is stored as sediment on the floor of the water body, which depends upon the rate at which organic matter is decomposed and deposited in said location.

**Mineralization** - The chemical transformation of compounds found in organic molecules into their inorganic counterparts, allowing for the ease of absorption on part of autotrophs.

**Closed Boundary Conditions** - In simulation, when an object is modeled, its exact shape can be emulated by the simulation software. This is known as open boundary conditions. However, the simulation is considered to use closed boundary conditions if it uses 1x1 squares (2D space), or 1x1x1 cubes (3D space) to model the object.

**Half-Saturation Constant** - The concentration of the substance in the liquid that allows for half the maximum uptake rate to be achieved. Useful for when dealing with smaller concentrations. Seeing as this is a partial rate, this involves the use expressions and equations with partial differentiation.

**Endogenous Respiration Constant** - When cellular material and/or overall biomass is oxidized as opposed to the nutrients they absorb to produce nutrients for growth.

**Limnology** - Study of the biology, chemistry, physical, and other aspects of a lake and other fresh water bodies.

**Diagenesis** - The chemical and physical modifications a decayed organic compound is to undergo before metamorphism from pressure and temperature (that is later on). This includes formation of minerals (e.g. calcium). What occurs in the process determine whether specimen remains in tact or not.

**Biocenoses** - Community of naturally-occurring organisms that cooperate with each other, are self-sufficient and strongly interlinked.

**Oligotrophic** - Reservoir/lake/similar ecosystem characterized by low nutrient concentrations and high DO at bottom levels. Mesotrophic, meanwhile, has intermediate amount of nutrients and decent productivity, but shows signs of water quality problems.

**Pisciculture** - Breeding and overall artificial control of fish populations.

**Cited references to follow up on**

Martinho, V., Monteiro, I.O., Fernandes, E.H.L., Janeiro, J., 2008. Numerical modeling of oil spills in the estuary of Patos Lagoon. Rio Oil and Gas Expo and Conference 2008. Rio de Janeiro. Proceedings of the Rio Oil and Gas Expo and Conference, vol. 1, pp. 1–8

Little, C., Kitching, J.A., 1996. The biology of rocky shores. In: Biology of Habitats. Oxford University Press. Mann, K., 1982.

Ecology of coastal waters – a systems approach. Studies in Ecology, vol. 8. Blackwell Scientific Publications, p. 322.

	Na, E.H., Park, S.S., 2006. A hydrodynamic and water quality modeling study of spatial and temporal patterns of phytoplankton growth in a stratified lake with buoyant incoming flow. <i>Ecological Modelling</i> 199, 298–314
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Does simulating bodies of water for biological parameters (e.g. DO, nitrates, etc.), using closed boundary conditions result in statistically significant differences in results compared to those produced when using open boundary conditions?</li> <li>2. How can nutrient transfer up the trophic pyramid be modeled using techniques similar to these ones (or other computational techniques), and what parameters, equations, and models would this involve?</li> <li>3. How can computational techniques such as the ones utilized here be used to simulate the relationship between different levels of iron fertilization and primary production in phytoplankton, and the ecological impacts of the iron?</li> <li>4. How major of a role do hydrodynamic factors influence phytoplankton populations in natural bodies of water compared to man-made bodies of water?</li> </ol>

## NOTES

### Abstract:

- Study took place in the Tucuruí reservoir, Pará State, Brazil, which is a dam located in Northern Brazil, just south of where the Amazon empties out into the Atlantic.
- Knowing the variables and relationships that influence water quality is important for managing local aquatic systems (lakes, ponds, reservoirs, dams, etc.)
- Simulations for high and low rainfall periods were performed using a Hydrodynamic Model (hereafter MOHID), a type of process-oriented modeling system
- Field Data Collected: Weather conditions including air temperature, humidity, wind, cloud cover, solar radiation; Marine conditions including DO, temperature, micronutrients including nitrates, ammonia, and phosphorus.
  - Collection occurred 1 July 2010 to 1 July 2011
- Simulation based on data collected at the inlets and outlets of the reservoir.
- Findings include that the reservoir has a limited amount of phosphorus, growth in blooms is impacted by varying concentrations of nutrients between high and low rainfall periods. Additionally, chlorophyll a is a key indicator for modeling the ecology of the reservoir, in particular with regards to algal blooms.
- This study helped develop a better understanding of the reservoir as a whole, and helped developing mitigation policies for algal blooms

### Introduction:

- As Brazil has developed and industrialized, it has made use of hydroelectric power for energy, but these systems have unknown and possibly irreversible consequences for marine ecosystems.
  - This is caused by the conflicting interactions of climate with the artificial currents generated by hydroelectric systems.
  - The Tucuruí reservoir (subject of the study) was one of the first hydroelectric power sources for the nation
- In particular, advective currents are modified by hydroelectric systems.

- Between light, temperature, and nutrients, nutrients are the only factor limiting phytoplankton growth that humans can (easily) control
  - This becomes important when considering the control of algal blooms, which is connected to nutrient concentrations
- However, in order for algal blooms to be regulated, it is important that the associated marine systems are well understood. Thus, this study aims to connect phytoplankton populations and nutrient concentrations to hydrodynamic forces.
- Most marine nutrients are lost due to uptake from algae. DO varies given the varying rates of photosynthesis (day) and respiration (night).
  - Greater loads of nutrient leading to greater uptake and oxygen production occurs during the day, transferring energy to higher-order organisms. Meanwhile, oxygen is depleted during the night, which can sometimes lead to anoxic conditions that harm higher-order organisms.
- Environmental processes can be better understood via mathematical modeling. Said models consider multiple factors including processes relating to water quality, sediment transport, and hydrodynamics. MOHID, a specific type of hydrodynamic model used in this field, which has these capabilities, is used in this study.
- Goal: Model the spatial and temporal trends of phytoplankton in the reservoir, connect phytoplankton populations and nutrient concentrations to hydrodynamic forces, as well as assess the impact on water quality (very multi-faceted).

#### Materials and Methods:

- Basic background information is provided about the reservoir,
  - Geographic coordinates (just south of the equator), surface area (2430 km<sup>2</sup>), water mean residence time (46 days), temperature range (27°C - 34°C), average stream flow (11k m<sup>3</sup>/s).
  - Since 1984, properties (same as ones mentioned in abstract) have been monitored.
  - Inflows from Araguaia, Tocantins, and Itacaiunas river (45%, 40%, and 5% contributions, respectively)
  - NE prevailing winds, October to April is when reservoir is most full
  - Reservoir has been studied through a variety of lenses (chemical, environmental, social, floral, faunal, economics, etc.), however, this study was first one to focus on water quality and hydrodynamics
- Provides background info on the MOHID model, and various math equations:
  - Portuguese model programmed using object-oriented programming in FORTRAN (formula translation) 95,
  - Contains various modules that are able to analyze physical and biogeochemical factors, including the hydrodynamic and water quality modules.
  - Hydrodynamic module uses the Navier–Stokes equations (see definition above)
  - Mathematical modeling of nutrient and organic matter cycling is based on conservation equations that take into account DO and dissolved nutrients in the pelagic and benthic phases, phytoplankton, zooplankton.
  - Equation used to model the primary production of phytoplankton, which is based off exponential growth models which measure the variation in biomass given initial biomass.

- Biomass multiplied by net primary production (Gross primary production minus respiration, grazing, mortality, and excretion)
- Use of maximum growth rate equation is used.
  - Maximum growth rate at a given temperature, multiplied by the limiting factors of temperature, light availability (as measured through radiation intensity), and nitrogen, ammonia, and phosphorus conditions
- Lambert–Beer law is used to model reduction in light availability given greater depth in the water body (i.e. light attenuation), and another equation is used for finding the water column light extinction coefficient, which is based off the phytoplankton self-shading effect, chlorophyll concentrations, and suspended particulate material.
- Implementing the Model
  - Bathymetric measurements of the reservoir were attained. Generally, the further south, the shallower the reservoir becomes.
  - Using the field data collected from July 2010-2011, the hydrodynamic and water quality models were set up and verified.
    - The following variables were inputted into the model: Bathymetry, air temperature, solar radiation (i.e. light availability), wind speed and direction, inflow and outflow, water temperature, concentrations of the various nutrients, DO, and total suspended solids.
    - Various parameters were instituted for the model. Their values were derived from the literature, and then adjusted so that the model results and field data could be in agreement.
    - Parameters included the following:
      - Maximum gross growth rate at the reference temperature
      - Optimum light intensity for phytoplankton photosynthesis
      - Nitrogen and Phosphorus half-saturation constants
      - Endogenous respiration and Excretion constants
      - Maximum mortality rate (at reference temperature)
      - Phytoplankton mortality half-saturation rate
      - Phytoplankton assimilation efficiency
      - Phytoplankton nitrogen/carbon and phosphorus/carbon ratio (redfield ratio)
      - Particulate organic nitrogen and phosphorus decomposition rates (at reference temperature)
      - Dissolved organic nitrogen and phosphorus mineralization rates (“”)
      - Nitrification and Denitrification (“”)
      - Oxygen/nitrogen ratio in nitrates, oxygen/phosphorus ratio in phosphates
- Statistics
  - Uses  $R^2$
  - Uses Root Mean Square Error (RMSE), which is like a population standard deviation, but for a time series. Essentially, instead of subtracting

the mean from each index, each predicted value is subtracted from the observed value.

- From there, the Normalized Objective Function (NOF), was used. This was equal to RMSE divided by the mean observed of the observed values. NOF values between 0.0 and 1.0 indicate ideal model calibration.
- Made use of special scatterplots, where the predicted values from the model were on the dependent axis, and the actual values observed were on the independent axis. The slope, gamma, indicates the error of the model, with 1 being perfect prediction. Thus,  $R^2$  and gamma closer to 1 should indicate that the model is well-calibrated.
- Data collected included:
  - Comparisons between model predictions and actual field data, specifically comparing temperature and depth measurements between model predictions and actual field data, both in the high and low rainfall seasons.
  - Comparisons between model predictions and actual field data for all the data collected at surface level of the reservoir
    - One at station M1, another at station M3
  - Comparisons between model predictions and actual field data for all the data collected at bottom level of the reservoir
    - One at station M1, another at station M3

#### Results:

- For hydrodynamic module, there is an overall good agreement:
  - There was a good match between the model predictions and field data for surface elevation of the reservoir.
  - The comparisons of temperature along the vertical profile between the model and the field data during both the wet and dry seasons showed pretty good agreements (see analysis of Figure 7/Table 2 above)
  - Also found that the circulation in the reservoir is mainly driven by wind
- Results for water quality module are likewise in good agreement:
  - Most gamma and  $R^2$  are within 0.2 or 0.3 of 1, which indicates that the models are fairly accurate, and that there is little straying of the actual field data from the predictions.
  - Similarly, most of the NOF values were within 0.5 of 0 (and 0.0 and 1.0 indicates proper calibration).
  - See analysis of figure 12 and table 3 above for more details.
- With the model validated, spatial and temporal variables for water quality, e.g. DO, chlorophyll and micronutrient concentrations, and their impact on phytoplankton were able to modeled (study provides multiple maps showing various color gradients to depict these trends. None of them are analyze here. Rather, the important findings derived from them are summarized in the notes below):
  - For phosphorus, during the dry season, upstream, there were lower concentrations (little inflow), but downstream, there were higher concentrations (urban dumping)
  - However, during wet season, the opposite is the case for each part of reservoir.

- All nitrate concentrations are found to be below 10 mg/L during the dry season, and during the wet season, there are more nitrates overall: <10 mg/L downstream, >30 mg/L upstream
- For ammonia, in both wet and dry seasons, downstream, concentrations were <0.04 mg/L, and upstream, >0.13 mg/L.
- DO in dry season, is 6-7.8 mg/L downstream, and 3-5.5 mg/L upstream. >7.8 mg/L and 3-5 mg/L respectively for the wet season
- For chlorophyll a concentration, model assumes that phytoplankton carbon concentration is directly proportional to concentration of the pigment in the water
  - Conversion factor of 60 mg C/mg chlorophyll was used. During both seasons, no concentrations exceeding 0.003 mg/L
- Article then proceeds to list out data from Tables 2 and 3, that we've already covered in figures section above.

#### Discussion:

- With model now validated, its results can be analyzed.
- Feb-Aug, the wet season provokes greater amount of mixing, less stratification (i.e. autumn/winter of the Southern hemisphere), and stratification is more in the hot, dry season (summer)
  - Trends we've observed time and time again.
- During thermal stratification, resources are released from the bottom layers.
- Analyzing wind velocities was prereq to analyzing impact of nutrients on phytoplankton
- In bottom layer, DO was higher during the dry season. Wind and rain lead to more mixing during wet season (plus more DO used by organisms), meaning DO is more uniform, less amount at the bottom.
- This possibly rings about anoxic conditions, harmful to the fish species of reservoir
- Station M3, which is shallower and hence always mixed, showed that at both bottom and top of water column, water properties were homogenous.
- Chlorophyll a concentrations were low b/c low water residence time inhibits algal growth
- Nutrients enter into the reservoir upstream being concentrated and saturated, and are then diluted downstream.
- Phosphorus conditions sometimes reach the legal limit. However, law does not account for trophic status of water, so other definitions were used for measuring this metric
- Nitrates are higher during wet season due to agricultural dumping. Upstream watershed transports nutrients. Law states that 10 mg/L means contamination, however, more scientific definitions considers 5-50 mg/L to be eutrophicated system.
- Upstream higher ammonia brought about by greater amounts of discharge in that area. No contamination on part of ammonia (ammonium?) observed, although its levels do indicate that the reservoir is mesotrophic.
- DO higher downstream b/c photosynthesis and reaeration, Lower DO upstream b/c decomposition, respiration, and reaeration. However, DO levels found indicate contamination for being below legal limit.
- By contrast, findings for chlorophyll a concentration don't show this to be the case, because it does not exceed 0.1 mg/L limit.
- N:P ratio of >10 indicates a phosphorus limited system. Seeing as here N:P = 100, the reservoir is undoubtedly phosphorus limited.

- This means management strategies must be focused on regulating P levels. However, P concentration might be independent of phytoplankton growth, meaning the system remains oligotrophic and has low chlorophyll a levels.
  - This indicates that reservoir water is quickly flushed out by the high incoming water flow. This means greater source of upward movement of nutrients came from cell lysis in the phytoplankton.
- We can safely assume the model is valid because  $\gamma$  and  $R^2$  values are close to 1, and NOF is close to 0.

#### Conclusion:

- Hydrodynamic module produced spatial and temporal distributions for the temperatures at varying points along the vertical profile, as well as the impacts of wind on the surface of the reservoir, during both wet and dry season
- Water quality module produced information about DO, chlorophyll a, various micronutrients.
- Although findings show that the reservoir is phosphorus-limited, there is a decreasing chlorophyll a gradient brought about by rapid inflow and outflow of reservoir, which inhibits phytoplankton growth and shows that hydrodynamics play a role in impact phytoplankton populations and eutrophication factors. (?)
  - So (i guess), even though, with phosphorus-limited environment, that is not what limits growth, rather the low amounts of chlorophyll a, which indicate lower primary production and productivity in the phytoplankton brought about the fast inflow and outflow, is what limits growth.
  - Nutrients are upwardly transported via cell lysis in the lower-level phytoplankton populations (?)
  - However, article suggest for future studies, analyzing relationship between chlorophyll a and eutrophication.
- Overall, this study has helped develop a useful tool for modeling various factors that impact phytoplankton populations, and can serve as an important tool for managing environmental conditions in water bodies (such as the reservoir).

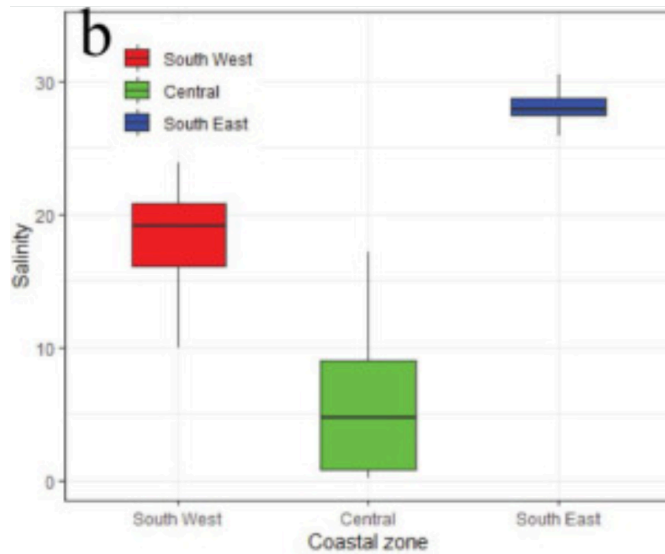


## Article #11 Notes: Environmental Controls of phytoplankton in the river dominated sub-tropical coastal ecosystem of Bangladesh

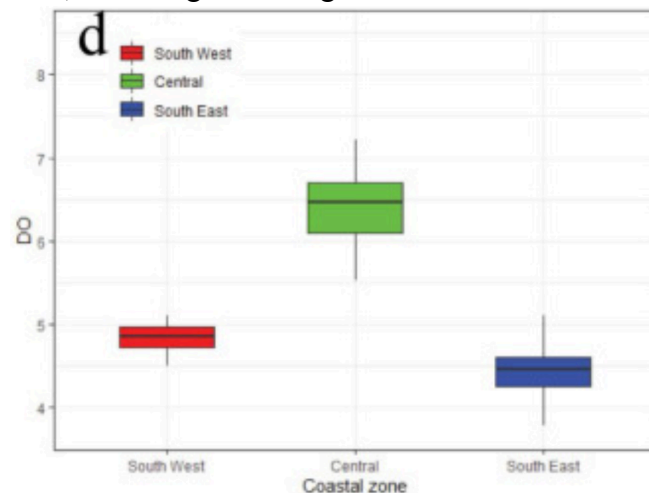
<b>Source Title</b>	Environmental Controls of phytoplankton in the river dominated sub-tropical coastal ecosystem of Bangladesh
<b>Source citation (APA Format)</b>	Sarker, S., Haque, A. B., Chowdhury, G. W., & Huda, A. N. M. S. (2023). Environmental controls of phytoplankton in the river dominated sub-tropical coastal ecosystem of Bangladesh. <i>Regional Studies in Marine Science</i> , 66, 103114. <a href="https://doi.org/10.1016/j.rsma.2023.103114">https://doi.org/10.1016/j.rsma.2023.103114</a>
<b>Original URL</b>	<a href="https://www.sciencedirect.com/science/article/abs/pii/S2352485523003043">https://www.sciencedirect.com/science/article/abs/pii/S2352485523003043</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Plankton, Ecology, Coastal ecosystem, Multivariate statistics, Northern Bay of Bengal
<b>#Tags</b>	<a href="#">Advanced Statistical Tests and Techniques</a> ; <a href="#">Drivers of Phytoplankton Dynamics Globally</a>
<b>Summary of key points + notes (include methodology)</b>	<p>Due to a severe lack of knowledge of marine conditions along the Bangladeshi coast, data on the various biotic/abiotic factors (DO, salinity, micronutrients, chlorophyll, turbidity, and temperature), was collected, and their ability to explain their impact on phytoplankton dynamics was assessed. The coast was split into southwest, central, and southeast, and using various instruments, the data were collected during northeast monsoon seasons of 2020, 2021, and 2022. After performing a series of advanced statistical techniques, the authors were able to conclude that turbidity, salinity, and silicates were major drivers of phytoplankton dynamics and overall coastal heterogeneity. It was also found that 44% to 73% of phytoplankton dynamics, namely abundance and spatial variability, can be explained by these variables, with salinity, micronutrients, turbidity and temperature (in that order) exerting impacts on phytoplankton. Valuable data on habitable ranges were provided. The article places these findings in broader ecological context: the southwest was found to be rich in nutrients due to nutrient cycling of mangrove forests and only moderately turbid and saline due to only moderate discharge, the central region rich in silicate and very fresh and turbid due to very high amounts of discharge, and the southeast very saline and nutrient deficient due to lack of discharge. From there, it draws connections to ecological niches of the different phytoplankton genera. While these data have many implications for conservation efforts, fisheries and simulatative capabilities and reflect the ecology of this region, a major limitation was that data was only collected during northeast monsoon. Further research into phenological and ecological factors is necessary.</p> <p>(overall, major stepping stone into data collection and trend analysis w/ phytoplankton, given multiple parameters! Although not the typical <math>x \rightarrow y</math> experiment, just note that)</p>
<b>Research Question/Problem</b>	How do the various regions of the Bangladeshi coast compare with regards to biotic and abiotic factors (DO, chlorophyll, temperature, micronutrients, turbidity, salinity, etc.), and in what nature

/ Need

does this drive phytoplankton dynamics?

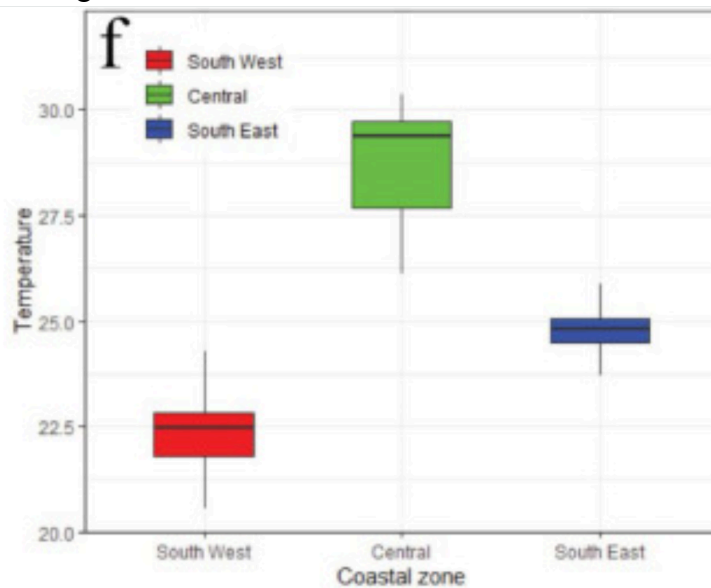
Important  
Figures

This figure illustrates the spread of measurements of salinity among the three regions (PSU). The data clearly indicate that the southeast region is the most saline, followed by the southwest and central regions. The median salinity for the central region is 5 PSU. The data is skewed towards higher salinity values, its upper tail of its distribution coinciding with the lower tail of the southwest region. The southwest's salinity distribution is skewed towards lower salinity, with a median of 19 PSU. Meanwhile, the entire distribution for salinity in the Southeast distribution is above those of the other two regions. It has a fairly symmetric distribution, with a median salinity at around 28 PSU, with range covering about 26 to 31 PSU.

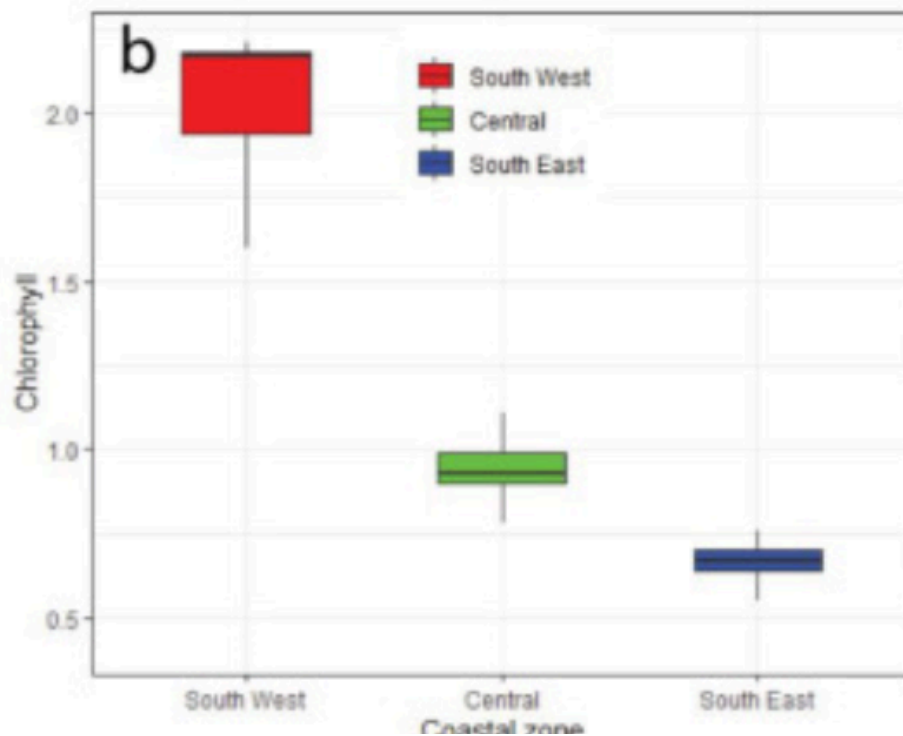


This figure depicts the range of DO across the three regions (mg/L). The data suggest that the central has the most varied but greatest DO, with the Southwest region having the most after that but the least variation, and southeast region being second most varied but having least DO. All distributions are relatively symmetric. Median DO values are (all values are approximate) 6.5, 4.8 and 4.5 mg/L, with ranges between 5.5 to 7.3, 4.5 to 5.2 and 3.8 to 5.1 mg/L for central, southwest, and southeast regions respectively. Distribution of DO for central region is entirely above the other

two regions.

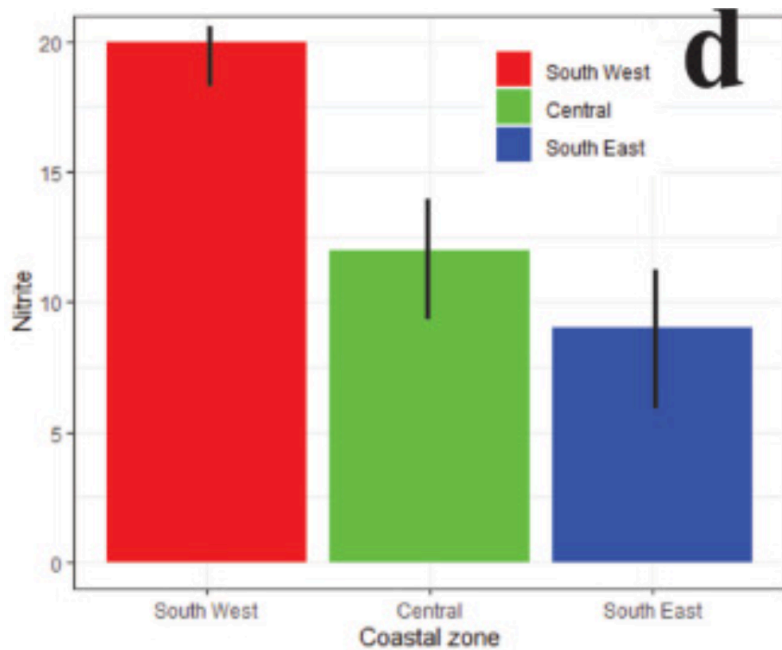
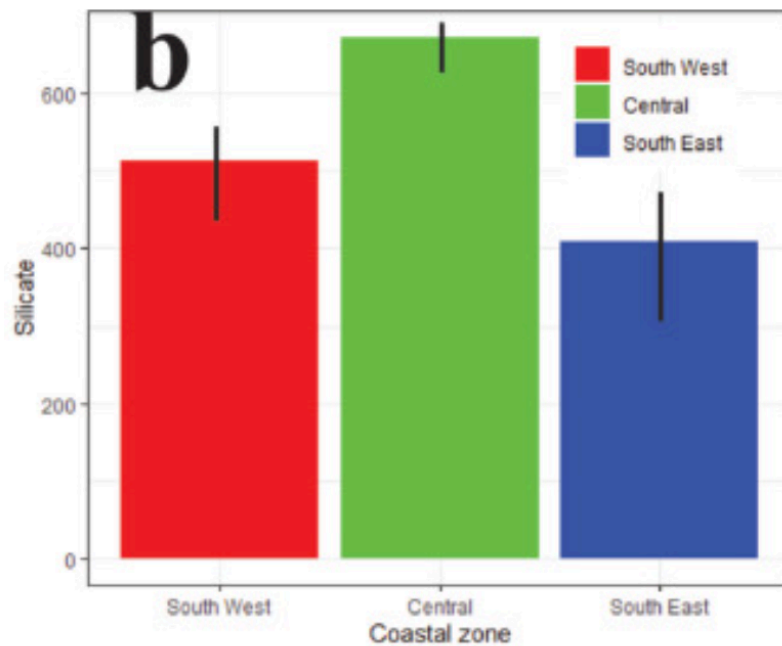


This figure depicts the range of water temperature across the three regions (°C). Overall, the data indicate that the central region is the warmest, followed by the southeast and southwest regions. The central region has a severe skew towards cooler temperature, although the entire distribution is greater than the other two regions. Median temperature is (all stated values are approximate) 29.4, ranging between 26.25 and about 30.3. Meanwhile, for the southeast and southwest regions respectively, median temperatures are 24.9 and 22.5, with ranges between 23.75 to 26.1 and 21.3 to 24.1.



These boxplots illustrate chlorophyll levels among the three regions (mg/m<sup>3</sup>). All distributions of

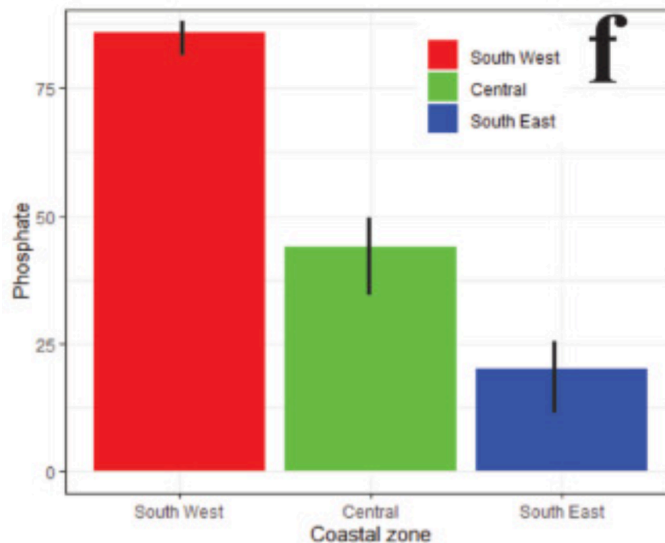
chlorophyll do not overlap with each other. This solidifies the assertion that the Southwest region, followed by the central and southeast, has the highest concentration of chlorophyll, meaning chlorophyll decreases west to east. The southwest distribution is extremely skewed towards lower concentrations, ranging from (all stated values are approximate) 1.63 to 2.20, with a median of 2.15. Meanwhile, the other two regions are fairly symmetric. Concentrations range between 0.77 to 1.13 with a median of 0.9 for the central region. In the southeast region, the range is between 0.55 and 0.75, with a median of 0.67.



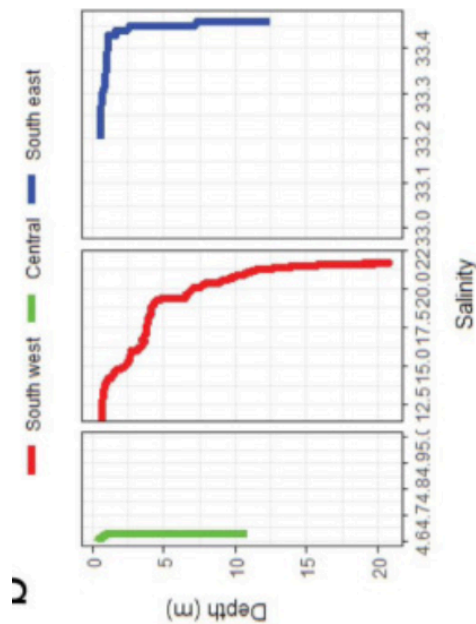
The bar graph at the top displays silicate concentrations, while the bottom one shows nitrate concentrations (mg/L - same for phosphates). The black lines represent the interval of values for

measurements across the three regions. It is apparent that the central region has the highest silicate concentration, followed by the southwest and southeast. Variation in concentration also increases in the same sequence. For the central, southwest, and southeast regions respectively, (all stated values are approximate), estimated concentrations were 675, 510, and 405, with ranges between 650 to 690, 440 to 560, and 315 to 475.

Meanwhile, nitrate concentrations were a lot lower, overall, given values for the concentrations among the three regions. From west to east, nitrate concentrations tend to decrease, while variability increases. From west to east, ranges in nitrate concentration are as follows (all values here are approximate): 18.5 to 20.3; 9.8 to 14.3; 5.3 to 11.3, with point estimates at 20.0, 12.4, and 9.1.

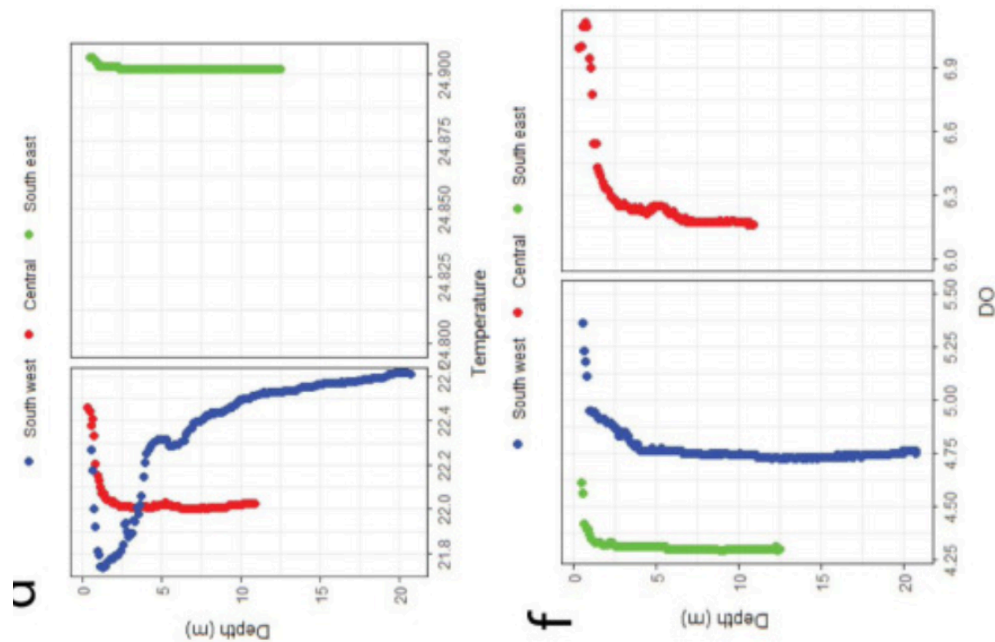


Similar west-east gradient can be observed for phosphates, although overall concentrations are higher than nitrates (but nowhere near silicates). From west to east, point estimates for phosphate concentrations are 86, 43, and 20, with ranges being between 80-87.5, 35-50, and 12-25.



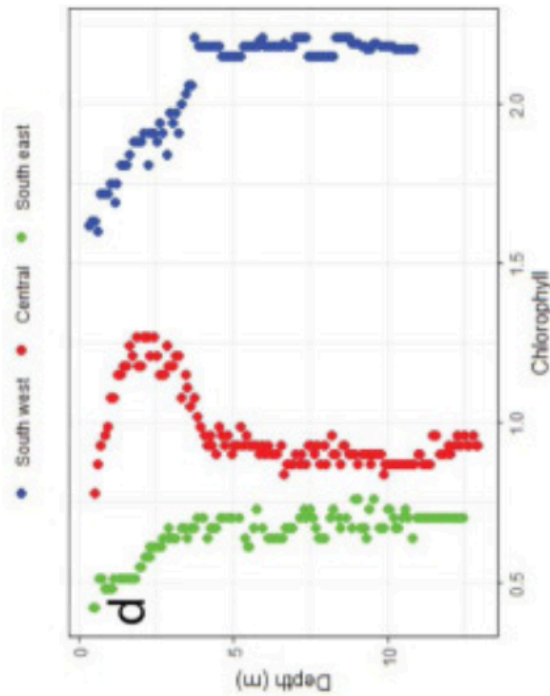
This dotplot describes the relationship between depth and salinity among the three regions. The order of appearance for each region on the graph is based on the ascending value of a given variable (in this case salinity) across the regions (as found and described in the above figures). Also, (and this goes for figures below), the original figure has been rotated counter-clockwise  $90^\circ$  in order to better analyze the value of the given variable, given depth. That being said, here is analysis for salinity (source of bias from the varying depths collected for each region):

For all regions, there seems to be a positive, somewhat logarithmic curve, that characterizes the relationship between depth and salinity. The salinity levels off quickest in the low-saline central region almost immediately, at around 4.65 PSU. In the more saline southeast, the salinity increases rapidly within about the first 2 meters, going from 33.0 to 33.5 before leveling off. Lastly, for the southwest region, has the most gradual gradient for salinity, leveling off only after between 10 to 15 meters of depth at about 21 PSU.

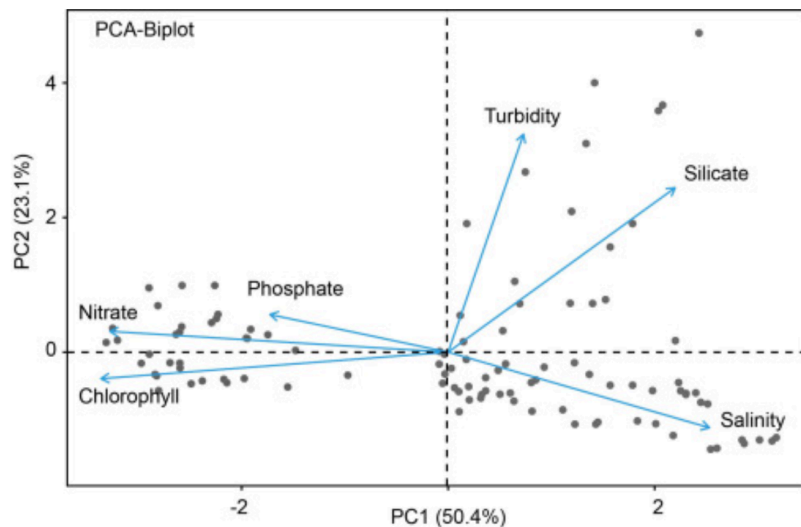


On the left, the relationship between depth and temperature is shown. In the warmer southeast region, the temperature remains about the same throughout all depths (although there is a slight decrease) at around 24.9°C. Meanwhile, in the central region, temperature rapidly decreases within the first 2.5 meters of depth, going from 22.5°C to 22°C before leveling off. Lastly, the most dynamic relationship can be observed in the southwest region: within the first 5 meters, the temperature decreases from 22.2°C to 22°C, before increasing back up to that value. Then, over the course of the next fifteen meters, the temperature increases, reaching about 22.6°C.

On the right, the relationship between depth and DO is shown. The data indicate there is a curve of exponential decay encapsulating this relationship for all regions. The (comparatively) anoxic southeast has the sharpest decline, going from 4.625 to 4.25 mg/L within the 2.5 meters. The decline is greater but more gradual for the southeast region, going from 5.38 to 4.75 within the first five meters before leveling off. The decline is yet greater and more gradual for the (comparatively) oxygen-rich central region, which, over the first 7.5 meters, declines from 7.05 to 6.15, leveling off from there.



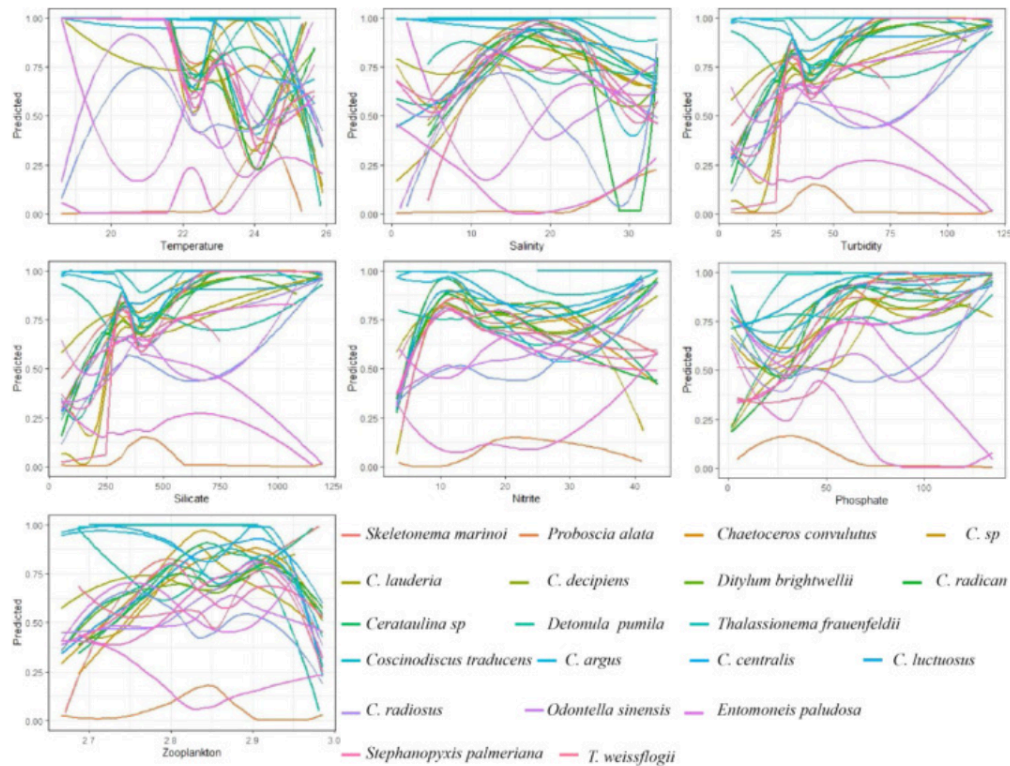
This scatterplot displays the relationship between depth and chlorophyll levels among the three regions. In the southeast, there is a small, gradual increase that levels off: concentration starts off at  $0.5 \text{ mg/m}^3$ , reaching  $0.75 \text{ mg/m}^3$  by five meters of depth, and leveling off. In the central region, within the first 3 to 4 meters, there is a negative concavity, with chlorophyll levels increasing from  $0.75$ , peaking at  $1.25$  around two meters deep, before decreasing and leveling off between  $0.8$  and  $0.9$ . Meanwhile, the southwest region has a positive linear relationship between depth and chlorophyll levels for the first four meters of depth, then levels off at  $2.2 \text{ mg/m}^3$ .



This graph illustrates the PCA of the environmental variables among the coastal regions. Larger magnitude vectors indicate that a variable is a more significant driver of the phytoplankton heterogeneity. Notice that nitrate, phosphate and chlorophyll have incredibly small vertical components, albeit large horizontal ones. This means they have smaller magnitudes and hence are



less impactful. Meanwhile, both the horizontal and vertical components of turbidity, silicate, and salinity are rather large. So, it is concluded that it is these factors that drive heterogeneity among coastal waters.



These are a set of AGAM functions depicting the impact of various biotic/abiotic factors on the 20 most abundant phytoplankton species among the three regions. The y-axis measures the abundance of the species in all cases. This has important ramifications on the habitable ranges of each species across the different variables.

Temperature:

Most species have a relatively high abundance, but at around 21.5°C, the abundance of the species begins to decline, reaching the lowest point around 24°C, before increasing once more, meanwhile other species continued to decrease in abundance. There are two notable species that have little abundance at lower temperatures, but greater amount at higher temperatures.

Salinity:

Discounting the one or two species that have no impact on abundance due to salinity, and the one or two species that prefer hypersaline/fresher environments, the habitable range of salinity for most species is between 15 to 25 PSU.

Turbidity:

With the exception of three species that seem to prefer less turbid (and thus more light-intensive) environments, it seems that after 25 FTU, the abundance of most of the other species increases, indicating that most species prefer an aquatic environment that is at least somewhat cloudy, so as to allow for excessive light intensity to be curtailed.

Silicate graph is the same as turbidity, which is problematic. See [below](#), that's the best possible analysis that can be offered.

Nitrates:

With the exception of one species that has very low abundance given the presence of any amount of nitrates, and another that prefers more extreme concentrations, there seems to exist a habitable zone between 8 to 32 mg/L for most species. From there, the abundance of certain species increases, while it decreases for others.

Phosphates:

With the exception of three species, whose abundance is negatively related to phosphorus levels, most species have an upward concavity in abundance between 0 and 50 mg/L, before stabilizing in a habitable zone for all concentrations thereafter.

Zooplankton:

One species seems to prefer extreme abundances of zooplankton. While one species has very low abundance given any presence of zooplankton, it confirms to the larger trend: Between concentrations of  $2.8$  to  $2.95 \times 10^3$  cells/L of zooplankton, there seems to exist a habitable zone.

**VOCAB:  
(w/definition)**

**Turbidity** - The level of clarity in water, with clearer water indicating lower turbidity.

**CTD** - In oceanography, CTD is an abbreviation for conductivity (i.e. salinity), temperature, and depth, and a common set of electronic equipment used to measure these properties.

**Spectrofluorometer** - An instrument that harnesses the fluorescence (ability to absorb higher frequency electromagnetic radiation and emit lower frequency electromagnetic radiation) of certain compounds of a sample to provide chemical information about that sample.

**Nansen (Niskin) Bottle** - A cylindrical tool used to measure water properties at specific depths without mixing in water from other depths.

**AutoAnalyzer** - A tool used to measure a fluid sample that is continually flowing via segmenting it using air bubbles.

**Molybdosilicate** - Standard way of determining silicate concentrations in water. The silica reacts with the molybdate to produce a yellow pigment, which, in addition to dilution, allows for the concentration to be determined.

**Colorimetry** - The measurement of a concentration of a chemical dissolved within a colored solvent. Colorimetry is the chemical analysis through use of pigments. This involves identifying differences in color within solvents and determining their values.

**Automated hydrazine reduction** - A common way of measuring nitrite and nitrate levels in water. One part of the sample undergoes a diazotization- coupling reaction, a complex organic chemistry reaction, which determines initial nitrite concentrations. Another portion of the sample reduces nitrate into nitrite, and then same reaction occurs. The yield of the original reaction minus this second reaction is equal to the concentration of nitrate in the sample.

**FTU (Formazine Turbidity Unit)** - Standardized way of measuring turbidity in water. Generally, turbidity is measured through analyzing how light is scattered. The light source can be visible light, UV, infrared, or other electromagnetic radiation. FTU is an umbrella term, and does not have a specific light source.

**Tukey's Post-hoc Test** - A follow-up to the ANOVA test (see also below: PERMANOVA), which determines if there is significance in any of the data overall are significant (i.e. are any one of the means significantly different from the rest), where the exact set(s) of means that are significantly different are determined.

**Non-metric Multidimensional Scaling** - A representation of the level of difference between the measure of two or more variables, that is then arranged in order of degree of difference.

	<p><b>Principle component analysis</b> - A method that reduces the multiple dimensions associated with a datapoint by eliminating some number of variables, allowing for easier analysis to be performed.</p> <p><b>Generalized Additive Model (GAM)</b> - A regression model independent of parameters that makes use of a smoothing function (a function that best fits the data and can best describe its patterns) to describe a relationship.</p> <p><b>Variance Inflation Factor</b> - A measure of how much the variation observed in one independent variable is due to its relationship/connections with other independent variables. If <math>VIF &gt; 2</math> is false, then there is no collinearity between two independent variables.</p> <p><b>Temperature-Salinity diagram</b> - A graph where predicted temperature is graphed against salinity as a means to define the profile of a water mass.</p>
<p><b>Cited references to follow up on</b></p>	<p>Xia, S., Gao, B., Fu, J., Xiong, J., Zhang, C., 2018. Production of fucoxanthin, chrysolaminarin, and eicosapentaenoic acid by <i>Odontella aurita</i> under different nitrogen supply regimes. <i>J. Biosci. Bioeng.</i> 126, 723–729.</p> <p>Silva, L.H.S., Huszar, V.L.M., Marinho, M.M., Rangel, L.M., Brasil, J., Domingues, C.D., et al., 2014. Drivers of phytoplankton, bacterioplankton, and zooplankton carbon biomass in tropical hydroelectric reservoirs. <i>Limnologia</i> 48, 1–10, <a href="https://doi.org/10.1016/j.limno.2014.04.004">https://doi.org/10.1016/j.limno.2014.04.004</a>.</p> <p>Henson, S.A., Cael, B.B., Allen, S.R., Dutkiewicz, S., 2021. Future phytoplankton diversity in a changing climate. <i>Nature Commun.</i> 12 (1), 5372. <a href="http://dx.doi.org/10.1038/s41467-021-25699-w">http://dx.doi.org/10.1038/s41467-021-25699-w</a>.</p> <p>Litchman, E., de Tezanos Pinto, P., Edwards, K.F., Klausmeier, C.A., Kremer, C.T., Thomas, M.K., 2015. Global biogeochemical impacts of phytoplankton: a trait-based perspective. <i>J. Ecol.</i> 103 (6), 1384–1396, <a href="https://doi.org/10.1111/1365-2745.12438">https://doi.org/10.1111/1365-2745.12438</a></p>
<p><b>Follow up Questions</b></p>	<ol style="list-style-type: none"> <li>1. How do abiotic and biotic factors vary across different portions of a river, how does the impact phytoplankton dynamics, and how can computational models take that into account?</li> <li>2. How do varying levels of DO impact phytoplankton dynamics across different species?</li> <li>3. What impact do the variables studied here (and other ones) have on migration patterns in phytoplankton?</li> <li>4. How do ecological niches of different phytoplankton species influence their response to varying levels of pertinent abiotic/biotic factors?</li> <li>5. How can the ramifications the impact of various biotic/abiotic factors on phytoplankton dynamics has for higher-order organisms be computationally modeled?</li> <li>6. How do meteorological events impact oceanographical conditions?</li> </ol>

## NOTES

### Abstract:

- Data collection in species-rich coastal subtropical regions has been a challenge, hindering development of conservation strategies and overall understanding of these environments
- Focus was on getting the raw variability in environmental conditions, relationships between these variables and phytoplankton dynamics.
- Data was collected from the coastal regions of Bangladesh during the monsoon seasons between 2020 and 2022.

- Following variables were researched:
  - Temperature
  - Salinity
  - Dissolved Oxygen
  - Chlorophyll a (referred to as just chlorophyll for notes on this article)
  - Turbidity
  - Micronutrients:
    - Nitrates
    - Phosphates
    - Silicates
- Findings indicate that these metrics vary radically across the subtropical ecosystems of coastal Bangladesh, with micronutrients, salinity, chlorophyll, and turbidity being major drivers of this variability. All of the variables explained 44% - 73% variability in phytoplankton species abundance
  - Southwest: Moderate turbidity and salinity, higher levels of nitrate/phosphate
  - Central: Low salinity, high turbidity and silicate concentration
  - Southeast: High salinity, low nutrient concentration
- 121 Phytoplankton species id'd. 66 occur in all 3 of the regions. Genera by region:
  - Southwest: *Coscinodiscus*, *Odontella*, *Nitzchia*, *Proboscia*, *Ditylum* and *Thalassionema*
  - Central: *Coscinodiscus*, *Odontella* and *Nitzchia*
  - Southeast: *Skeletonema*, *Ditylum* and *Proboscia*

#### Introduction:

- Base of marine food webs (note specifically pelagic)
- Not just carbon cycle and climate regulation, also regulates nitrogen, silica, and phosphorus
- Various abiotic and biotic factors impact taxonomic composition, biomass, spatial distribution of species, and other fundamental phytoplanktonic dynamics.
- Background on Bangladeshi geography
  - Lots of rivers, major deltas (ganges, brahmaputra, etc.) emptying out into Indian Ocean, subtropical monsoon climate
  - $3.2E4 \text{ m}^3$  of freshwater and  $\sim 10^3$  of sediments discharged (4th and 1st largest globally, respectively), which plays major role in regulating coastal ecosystems, which is a major source of food and economic livelihood for the nation
    - Fisheries rely on phytoplankton, whose dynamics are changing
- Therefore, it is important to study phytoplankton on Bangladeshi coast, considering large geographic area and many explanatory variables, which hasn't been done previously.
  - This hinders understanding and thus conservation efforts of aquatic ecosystems. Therefore major goal is to minimize data gaps for this major geographic area.
  - Specifically, studying spatial variability of coastal Bangladesh, looking at the impact of the various physical/chemical variables (specified above) on phytoplankton

#### Materials and Methods:

##### Measurement:

- Data collection split into 3 zones: southwest, central, and southeast. (hereafter SW, C and SE) High sample size collected for each region, which spans 710 km coastline, and 21-23 °N/ 89-93 °E
- Samples collected during high tide, which occurred 2x (semi-diurnal), 3/4 m amplitude
- Data was collected during January and February of 2020-2022, as that is when the Northeast monsoon occurs.
  - Southeast monsoon season has waves that are too rough
- Given the large area the study was covering, rather than simultaneously collecting data from all stations, over cycle of 7 consecutive days, data from all stations was collected.
- Table I gives insight into measurement methodology:
  - CTD and mini-CTD were used to measure temperature and salinity. CTD alone was used to measure DO, and, in conjunction with spectrofluorometer, chlorophyll. Spectrofluorometer alone was used for turbidity, while Niskin bottle was used for the micronutrients. Plankton nets used to collect phytoplankton and zooplankton
  - Mini CTDs were located at 2500 sampling stations, CTDs and spectrofluorometers at 210, 130 for Niskin bottles, and 120 for plankton nets.
  - SW had 76 sampling stations for temperature, salinity, DO, chlorophyll, and turbidity; 40 for the micronutrients, 45 for both planktons
  - Chad 70 sampling stations for temperature, salinity, DO, chlorophyll, and turbidity; 50 for the micronutrients, 43 for both planktons
  - SE had 64 sampling stations for temperature, salinity, DO, chlorophyll, and turbidity; 40 for the micronutrients, 32 for both planktons
- Salinity, temperature, DO, turbidity and chlorophyll were measured on-site, while micronutrient concentrations and phytoplankton and zooplankton abundance were measured in lab.
- Mentions CTD (use described above), along with chlorophyll and DO sensors, are common tools for understanding all aspects of a water column, monitoring algal growth and distribution, and outlining water masses.
- Mini-CTD attached to was attached to the boat, measuring pelagic temperature and salinity in 1 minute interval, operating for about 2500 times (thus 2500 sampling stations as described above)
  - Mini-CTDs are also used in studying migration patterns in fish and other aquatic organisms.
- Quality of data was checked → made supplementary info., interesting...
- Fluorometer measured its variables (including green-blue algae unmentioned above) along the vertical profile, performing rapid measurement without preparing the samples.
- Water samples collected were put through a 0.45 µm cellulose-ester filter in order to measure/analyze nutrients.
- An AutoAnalyzer (def'd above, as a the three methods below) was then used for the micronutrients:
  - Molybdosilicate method used for silicates
  - Semi-automated colorimetry used for phosphates
  - Automated hydrazine reduction used for nitrates

- Phytoplankton and zooplankton caught wet plankton net, preserved with 3% formalin solution and analyzed under microscope in order to identify species (based off phenotypic analysis, referencing guides).

Statistics:

- First, the spatial variability of the data collected was modeled, then, heterogeneity was calculated, and the factors driving these conditions were also determined.
- Spatial variability mapped on ArcMap software, discrete color scale used for displaying values for the variables analyzed.
- Mean and SD of the variables studied were calculated, being normally distributed.
- One-way ANOVA test used to confirm presence of spatial variability for these variables.
- Tukey's post-hoc test was then used to see which groups were significantly different.
- Temperature-salinity diagram used for understanding overarching characteristics of this water mass.
- Vertical profiles of salinity, temp, DO, turbidity, and chlorophyll established, averages calculated.
- Heterogeneity was measured using Non-metric multidimensional scaling (NMDS) ordination and Principle component analysis (PCA). NMDS was able to collate samples with similar values, identifying similarities and differences among the data. NMDS analysis was then used to produce a graph depicting the overlapping of ordination.
- Permutational Multivariate ANOVA was used to find the statistical significance of the differences observed from the NMDS process, while PCA helped identify drivers of homogeneity.
- Generalized Additive Model (GAM) used to observe the role that these biotic/abiotic factors play in driving phytoplankton dynamics. The dependent variable was phytoplankton abundance across different species, based off independent variables of each environmental factor (separately)
- With so many variables creating complex relationships, it is possible that there is collinearity among the different variables, meaning that can just be disregarded as extraneous. In order to evaluate that, Variance Inflation Factor (VIF) was performed, eliminating any instances that resulted in  $VIF > 2$ .
  - (so goes the reasoning of the article, although personally I'm sure whether that's a wise decision)
- With that in mind, the following equation was established:
 
$$\log(N_{i,j}) = \delta + f_k(X_{i,k}) + \gamma$$
  - N and X are matrices. Matrix N contains rows i, which represents the sampling stations, and columns j, which are the each of the species. Matrix X contains rows i and columns k, the value of the given environmental factor. Lowercase delta is the intercept, lowercase gamma is unaccounted variation, log transformation is performed on the N matrix, and  $f_k$  represents the smoothing function performed on matrix X, given the explanatory variable analyzed.
  - Smoothing is determined by a restricted maximum likelihood approach, given that the function is transformed, both by the smoothing function and the logarithm.
  - GAM outputs were analyzed in order to develop a better picture of the impact of explanatory variables on phytoplankton dynamics.

Results:

- (See extensive analysis of figures above) Key findings/things to note:
  - Silicate > Phosphate > Nitrate
  - Nitrate, Phosphate and Chlorophyll decrease west to east
  - East is most saline.
  - DO, temp, silicate highest in central region.
- Salinity: ANOVA: Significant; Turkey Post-Hoc: central < other regions
- Temperature: ANOVA: No significant difference
- From T-S Diagram: Central: low salinity & density (meaning freshwater environment); Southeast: high salinity & density, Southwest moderate for both metrics.
- DO: ANOVA: Significant; Turkey Post-Hoc: central > other regions
- Micronutrients: ANOVA: Significant; Turkey Post-Hoc: Silicate in central significantly greater; nitrate/phosphate greater in southeast.
- Turbidity: Significant difference. 140, 100 and 60 FTU for central, SW, SE respectively.
- With regards to depth and the various variables, no significant differences **within** each coastal region, but the differences of each variables among the profiles of the different regions were significant.
  - Suggests that there is good mixing in the water (variation suggests stratification)
- Phytoplankton dynamics observed through the lens of zooplankton, phytoplankton community comp, and chlorophyll concentrations
- Higher chlorophyll in SW suggests that it is likely to be productive (which seems to be true, given phytoplankton biomass and diversity measurements below):
- Chlorophyll was uniform within regions, but varied significantly between them; ANOVA significant, and Turkey Post-Hoc SW > other regions to be significant.
- 121 species ID'd: 81, 71, 56 in SW, SE and central respectively, although diversity was calculated to be 2.58, 2.21, and 2.01 for SW, SE and central coasts.
  - 66 species across all regions (indicating wider niches), 43 not present in central region (indicating they are not fit for high-silicate/turbidity and fresh conditions), 13 not present in SE (indicating they are not for high-saline & phosphate/nitrate, low-moderate silicate/ turbidity)
- Average biomass across all regions:  $10.65 \times 10^3$  cells/L,  $\pm 2.66 \times 10^3$  cells/L. From SW to SE, biomass decreased. Similar observations for zooplankton
- In terms of heterogeneity, the NMDS performed indicated that the three regions were in fact distinct from each other.
- PERMANOVA tests further confirm this: SW different from Central w/o overlap, small overlap when compared to both central and SE.
- PCA shows that silicate, salinity & turbidity as major drivers of heterogeneity (see above)
- AGAM model used to assess impact of the 7 variables (micronutrients, zooplankton, turbidity, salinity, temperature) among the 121 phytoplankton species.
- Overall, these seven variables explain 44%-73% of the variation in phytoplankton biomass, taxon-comp (diversity), spatial variability (which gets into migration), etc.
  - Ofc, this is from looking at variation among the different species, which ofc varied (*Prorocentrum lima* best explained, *Actinellius* sp. worst)
- Phytoplankton spatial variability explained by variables in following proportions:
  - #1: Salinity (59.50% of species)
  - #2: Phosphate (10.74% of species)

- #3: Zooplankton (9.09% of species)
- #4: Silicate (8.26% of species)
- #5: Nitrate (7.43% of species)
- #6: Temperature (4.96% of species)
- #7: (turbidity not given)

*Interesting that they did not consider DO... will add as a Q, therefore.*

- Phytoplankton abundance explained by variables in following range of proportions (explaining factor of observed variation in abundance):
  - Salinity: 5%-28% *Tropidoneis* sp.
  - Phosphate: 8%-20% *Chaetoceros* spp.
  - Zooplankton: 5%-15% *Chaetoceros* spp.
  - Silicate: 7%-20% *Chaetoceros* spp.
  - Nitrate: 8%-24% *Pleurosigma* spp.
  - Temperature: 3%-5% *Thalassiosira* spp.
  - Turbidity: 2%-13% *Chaetoceros* spp.
- Additionally, each variable was the most impactful for given species/genera (listed ^^)
- See above analyzation of the AGAM model (very important).

Discussion:

- Spatial variability attributable to the variation in salinity brought about by variation in river discharge.
- Central region gets most river discharge (1.5 m<sup>3</sup>/s during SW monsoon, 61k during NE)
- SW: 30k m<sup>3</sup>/s during SW monsoon (>SE), and 100 m<sup>3</sup>/s during NE (<SE)
- SE: 5565.7 m<sup>3</sup>/s during SW monsoon (<SW), and 419.80 m<sup>3</sup>/s during NE (>SW)
- This explains the low-saline and high turbidity/silicate environment of central region. Exposure to coastal waters impacts SW and SE conditions.
- Conversely, the SE is high-saline and low nutrient due to less river outflow.
- SW: Nutrient cycling on part of mangrove plants + leaf litter, and moderate discharge (construction of barrage) makes it moderately saline/turbid, high nitrate/phosphates.
- DO, chlorophyll, salinity, density, and turbidity were uniform → this indicates vertical mixing, which is based off tidal range, water current and river discharge.
- Strong winds during monsoon season and other subtropical climatic events drive tidal range and water currents
- Mangrove forest ecosystems and their contributions of organic matter is why SW was most predisposed to productive ecosystem (rich nutrients and phytoplankton abundance/diversity). Central has less (though gets nutrients from big discharge), and SE the least b/c of less discharge.
- Diatoms are dominant in Bangladeshi coastal waters, which is related to the micronutrients studied (silica, nitrates and phosphates). Having access to these resources, they are the most abundant species.
- By region, in the center, *Coscinodiscus*, *Odontella* and *Nitzschia* genera were most abundant. C. b/c adapted to lack of light, cytological significance of silica. The other two likewise rely on the high silicate levels.
- *Skeletonema*, *Ditylum* and *Proboscia* genera most popular in the SE. This is due to the favorability of high salinity for these species, and their ability to tolerate lack of nutrients.



- Similarly, SW environment is ideal for following genera: *Oscinodiscus*, *Odontella*, *Nitzschia*, *Proboscia*, *Ditylum* and *Thalassionema*
- Salinity, micronutrients, zooplankton, water temperature, in that order, exerted a descending impact on phytoplankton dynamics.
  - Salinity/micronutrients has to do w/ varying river discharge/nutrient flux conditions. Along w/ turbidity, this is main driver of heterogeneity
  - Zooplankton control phytoplankton populations through feeding. They have a positive relationship with phytoplankton abundance.
  - Although temperature impacts metabolic rates, since temperature did not vary significantly for this study, it did not have that much of an impact on dynamics.
- Addresses how this varies globally!
  - Temperate: nutrients, light, zooplankton, temperature
  - Polar: Under-Ice light field, macronutrient concentrations
  - Tropical/Subtropical: pH, Salinity, nutrients from runoff, temperature, light (thus, findings are in line with what is regional norms)
- Heterogeneity means development of niches among phytoplankton species (adaptive radiation???), allowing for co-occurrence and biodiversity

#### Conclusion:

- Implications: Climate change impact on fisheries, conservation actions, general info for fisheries, simulation data (!!!), marine spatial plan (i.e. modelling)
- Major limitations: only looked at data from NE monsoon.
- Areas of future foci: Collecting monthly data to understand phenological factors impacting phytoplankton, other aspects of phytoplankton dynamics (competition, carrying capacity, etc.), analyzing niches of phytoplankton and more broadly, trophic/ecological ramifications

## Article #12 Notes: Incorporating carbon sequestration into lake management: A potential perspective on climate change

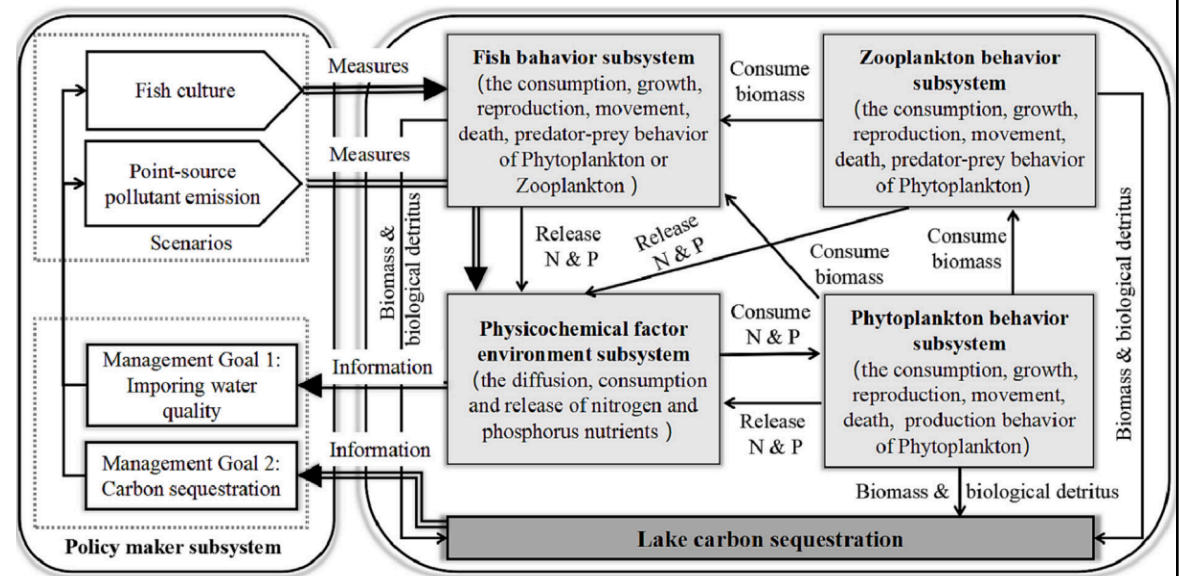
<b>Source Title</b>	Linking phytoplankton community metabolism to the individual size distribution
<b>Source citation (APA Format)</b>	Tian, Y., Zhao, Y., Zhang, X., Li, S., & Wu, H. (2023). Incorporating carbon sequestration into Lake Management: A potential perspective on climate change. (F. A. L. Pacheco, Ed.). <i>Science of The Total Environment</i> , 895, 164939. <a href="https://doi.org/10.1016/j.scitotenv.2023.164939">https://doi.org/10.1016/j.scitotenv.2023.164939</a>
<b>Original URL</b>	<a href="https://www.sciencedirect.com/science/article/pii/S0048969723035623">https://www.sciencedirect.com/science/article/pii/S0048969723035623</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Climate change, Lake management, Carbon sequestration, Water quality, Wuhan
<b>#Tags</b>	<a href="#">Potential Source of Raw, Actual Data on Environmental Factors Influencing Phytoplankton Dynamics</a> <a href="#">Searching for Factors Impacting Carbon Sequestration</a> <a href="#">Possible Biotechnological Connections</a>
<b>Summary of key points + notes (include methodology)</b>	In order to better understand the carbon sequestration capabilities of lakes and offer policymakers an important perspective on lake management, this study collected data of numerous parameters from 63 lakes in Wuhan, China in 2018, seeking to measure carbon sequestration capacity, driving parameters for chlorophyll concentrations in phytoplankton, and form policy proposals for lake management based off testing these key parameters in hypothetical scenarios. Another goal was to better establish the relationship between broader, ecological parameters, and smaller-scale biophysiochemical parameters. Through a vertically generalized production model, it was found that the average carbon sequestration capabilities of lakes in this region was $0.87 \text{ kg C/m}^2\text{a}^{-1}$ , which is significantly greater than forests and oceans. Next, a hierarchical linear model (HLM) was developed through a series of steps, including an unconditional means model to validate HLM use, a single- and multi-variable random coefficient model to identify and order level-one parameters that were drivers of chlorophyll concentrations (which indicated phytoplankton ability to sequester carbon). A mixed effect model was then implemented to identify significant level two parameters, and any relationship they held with level one parameters. Overall, it was found that phosphorus, nitrogen, <i>Rotifera</i> genus of zooplankton, and water temperature, in that order, were significant level one parameters. However, there no significant level two parameters, indicating that it is the deeper, biophysiochemical factors that are stronger drivers of phytoplankton dynamics. The only significant level one-level two relationship identified was a negative correlation between grassland and water temperature. Lastly, policy

reccomendations were developed through a multiagents-based model on the NetLogo software. Two tests were performed, one where pollutant (nitrogen and phosphorus) cocentration were controlled, and one where fish feedin was controlled. These tests found that phosphorus significantly increased phytoplankton sequestration capabilities, whereas nitrogen had little effect. Moreover, the phytoplankton were able to absrob pollutants, improving water quality while avoiding eutrophication. In the other experiment, it was found that exclusively zooplankton-feeding fish maximized carbon sequestration capabilities. Therefore, policy proposals were made to insett nitrogen and phosphorus into the lakes, while maintaing a tight control on their nutrient stoichiometry, and implement more fish that feed exclusively on zooplankton. This study is a perfect example of how parameters can be organized, and their relationships determined when it comes to phytoplankton dynamics. However, more work needs to be done in determining what factors influence what are the driving parameters and creating more precise policy reccomendations.

**Research Question/Problem/Need**

For lake ecosystems in particular, what carbon sequestration capabilities do they possess, what factors most influence phytoplankton dynamics, and in turn, these levels of carbon sequestration, and given this information, how can robust policy decisions be made to ensure propey water quality and enhance carbon sequestration among lakes (specifically in Wuhan, China for this study)?

**Important Figures**



This flowchart is an explanation of the lake-ecological-social management system. Starting from the scenarios section, we have modification to fish populations. From this, we get metrics on fish behavior, such as movement, role in the food chain, especially with regards to phytoplankton and consuming them (and zooplankton as well), among other ecological roles. These activity release phosphorus and nitrogen. Zooplankton play a similar role to fish in this flowchart, although they are intermediate group along the food chain. Phytoplankton both release and absorb P and N. Both zooplankton and

phytoplankton biomass and exportation allow for C seq. Additionally, pollutants as P and N to the system. This provides helpful information for understanding how to improve water quality and C seq.

**Table 2**

The estimation of fixed effects and random effects in Single RCM.

Model	Fixed effect (with robust standard errors)		
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (Temperature) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.29***	(7.46)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	1.21**	(0.46)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (TN) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.29***	(7.46)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	5.05***	(1.70)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (TP) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.29***	(7.46)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	47.55**	(19.26)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (TN:TP) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.29***	(7.46)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	-0.12	(0.09)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (Rotifera) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.29***	(7.46)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	2.45***	(0.63)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (Cladocera) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.72***	(7.53)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	0.41	(0.55)
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (Copepoda) + r_{ij}$			
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.38***	(7.52)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	0.04	(0.03)

\*, \*\* and \*\*\* indicate the significance at 10 %, 5 %, and 1 % levels, respectively.

This table illustrates relationship of various environmental parameters with chl a concentrations. The equations on the left represents the function used for the linear models for each of the relationships. Parentheses on the right indicate standard error. The parameter is plugged into the parentheses.  $\gamma_{00}$  is the intercept of each of the models.  $\gamma_{10}$  is the slope for each model. For example, the slope between temperature and chla is 1.21, meaning for every increase in temperature by one degree, 1.21  $\mu\text{g/L}$  chla would occur. Using that for nitrogen and phosphorus, for every increase in mg/L of N and P, there was 5.05  $\mu\text{g/L}$  and 47.55  $\mu\text{g/L}$  chla increase respectively. Similar idea of biomass of zooplankton. All other ones increase/decrease by an amount that is close 0, meaning that factor isn't very impactful. For every increase by 1 mg/L in ratio of P:N, there is only a 0.12  $\mu\text{g/L}$  chla decrease.

**Table 3**

The estimation of fixed effects and random effects in multivariate RCM.

	Fixed effect (with robust standard errors)		Random effect (with variance components)		
$Chl-a_{ij} = \beta_{0j} + \beta_{1j} * (Temperature) + \beta_{2j} * (TN) + \beta_{3j} * (TP) + \beta_{4j} * (Rotifera) + r_{ij}$					
$\beta_{0j} = \gamma_{00} + u_{0j}$	$\gamma_{00}$	93.30***	(7.46)	$r_{ij}(\sigma^2)$	74.60 (5565.10)
$\beta_{1j} = \gamma_{10}$	$\gamma_{10}$	1.27**	(0.56)	$u_{0j}(\tau)$	56.60 (3203.18)
$\beta_{2j} = \gamma_{20}$	$\gamma_{20}$	6.38**	(0.26)		
$\beta_{3j} = \gamma_{30}$	$\gamma_{30}$	6.82*	(28.97)		
$\beta_{4j} = \gamma_{40}$	$\gamma_{40}$	1.95***	(0.60)		

\*, \*\* and \*\*\* indicate the significance at 10 %, 5 %, and 1 % levels, respectively.

From the results in table II, we now have a multivariate RCM that compares the magnitude of the impact of each variable on chl a concentrations. Phosphorus is at  $\gamma =$

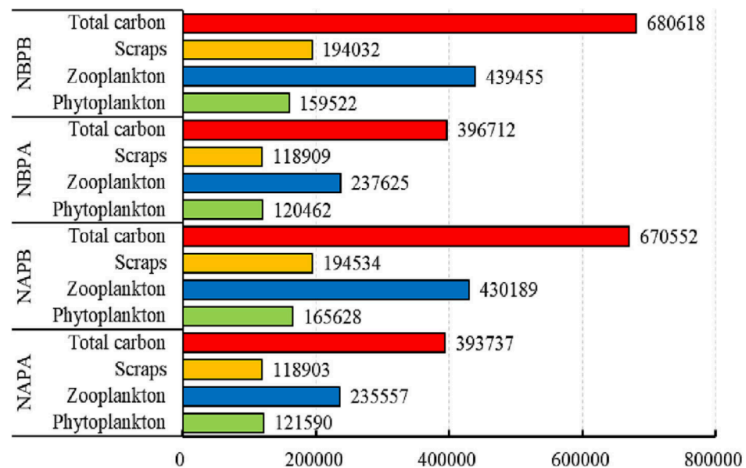
6.82, the highest amount of magnitude, but it is of the lowest significance, and note that its error is quite large. Following the decreasing  $\gamma$  values from there, nitrogen, then *Rotifera* biomass, then temperature have the next biggest impact on phytoplankton.

**Table 4**  
The estimation of fixed effects and random effects in MEM.

Models	MEM# with			
	Cultivated land	Forest land	Grassland	Artificial land
Fixed effects (with robust standard errors)				
$\gamma_{00}$	93.46***(7.33)	93.22***(7.27)	93.30***(7.46)	93.27***(7.47)
$\gamma^{10}$	1.35**(0.57)	1.22*(0.56)	1.16*(0.53)	1.28**(0.56)
$\gamma^{20}$	4.90*(2.76)	6.34***(2.72)	4.83(3.13)	5.11(3.19)
$\gamma^{30}$	3.45(26.22)	11.53(27.41)	10.26(31.47)	7.50(29.76)
$\gamma^{40}$	1.87****(0.64)	2.15****(0.68)	1.90****(0.61)	1.78****(0.65)
$\gamma_{01}$	54.24*(29.97)	-452.75***(214.83)	-82.49(324.96)	13.69(32.53)
$\gamma_{11}$	1.46(2.49)	-2.98(13.72)	-46.64***(19.97)	-1.62(2.78)
$\gamma_{21}$	-19.48 (12.33)	-36.82(0.57)	18.89(114.13)	10.21(11.63)
$\gamma_{31}$	107.39 (123.24)	772.85(0.31)	502.96(1127.71)	-57.62 (115.60)
$\gamma_{41}$	-1.99(2.21)	20.67(22.22)	-19.51(25.72)	2.20(1.90)
Random effects (variance components)				
$\tau_{ij}$	74.34	74.59	74.23	74.54
$(\sigma^2)$	(5526.27)	(5562.97)	(5509.96)	(5556.87)
$u_{0j}$	55.85***	55.56***	57.15***	57.04***
$(\tau)$	(3119.74)	(3086.70)	(3265.97)	(3253.39)

\*, \*\* and \*\*\* indicate the significance at 10 %, 5 %, and 1 % levels, respectively.

Adding on the level 2 parameters in order to establish the MEM, we see that only one combination has any significance: chl a is decreased because there is a negative relationship between the grassland and temperature. The grassland environment changes the temperature such that chl a concentrations decline. Otherwise, there is nothing significant.



**Fig. 8.** Maximum carbon value within 600 ticks under the point-source pollutant emission change scenario. In the initial water environment, the nitrogen and phosphorus concentrations were 1.5 mg/L and 1.0 mg/L, respectively, and the water temperature was 20 °C.

This bar graph indicates C seq given varying pollutant (N/P) phosphorus conditions). Given the description of the experimental groups, the data clearly indicate that higher amounts of phosphorus allow for the increase in C seq, and that nitrogen and C seq seem to be ambivalent to each other. Consider that NAPA and NBPA are nearly equivalent in C seq, 393,737 versus 396,712. Similarly, C seq in NAPB and NBPB is 670552 and

680618, respectively. When PA goes to PB, it is significant change, increasing by about 73%, but the values do not change much when N increases.

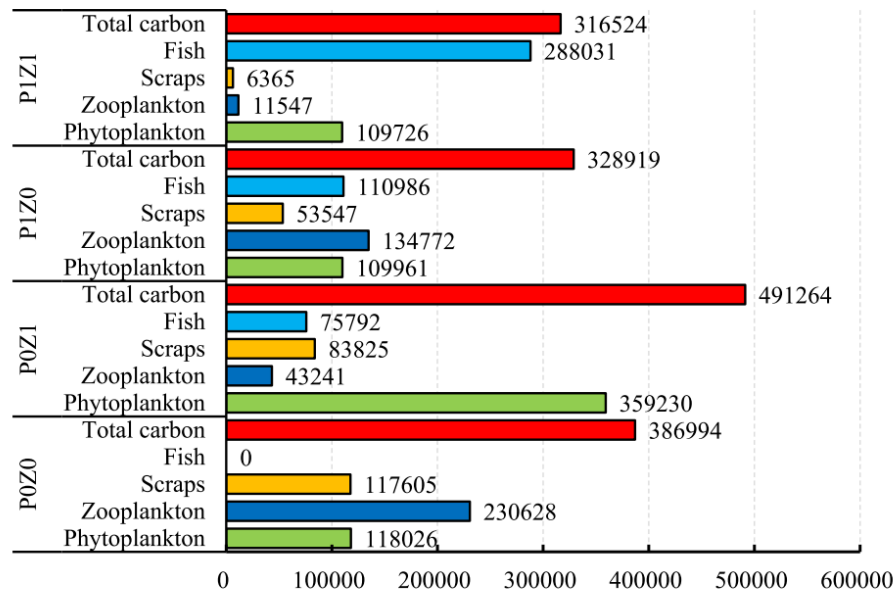


Fig. 9. Maximum carbon value within 600 ticks under the fish culture change scenario.

This figure illustrates the impact of different fish culture conditions on C seq. Relative to the control, the data indicate that any presence of phytoplankton-feeding fish decreases C seq, and that zooplankton-feeding fish increase C seq. This is because, given the experimental design description below, the control is 386,994, and the other two groups with P1 have 328,919 and 316,524. Meanwhile, the exclusive zooplankton-feeding group has a value of 491,264 (27% increase).

**VOCAB:**  
(w/definition)

**Tick** - A unit of time that is used by internal computing systems.

**Integrated method framework** - A methodology that entails evaluating a model through a multidimensional lens, analyzing all of its components, and seeing its performance in predicted scenarios as well as comparisons to real data.

**Redundancy Analysis (RDA)** - A method of data analysis that acts as an extension of multiple regression whereby the results/variation in a set of dependent variables is linked to a set of independent variables via the use of matrices. It is a sort of restricted PCA as only one possible ordination for both x and y are created.

**Canonical correspondence analysis** - A means by which parameters driving ecological conditions can be extracted from multivariate relationships, in turn elucidating relationship between species and their biological and ecological systems.

**Chemical oxygen demand** - Indicator for how much oxygen is present that can be used for carrying out chemical reactions, specifically oxidizing inorganic and organic molecules for nutrients.

**Permanganate index** - A means through which water quality is assessed. Potassium permanganate is boiled under acidic conditions, and oxidation is used to determine the index value.

**Unconditional means model** - A sort of null hypothesis used at for HLM in two-level

	<p>models. Sees if given IDV at one level significantly impacts DV at the other.</p> <p><b>Random coefficient model</b> - A special type of mixed linear model that can be used when there are repeated x/y measurements on the same subjects (b/c we have multiple variables we're analyzing). The mixed model has both the fixed and random effects.</p> <p><b>Mixed effect model</b> - Indicates the degree to which level 2 variables in HLM impact the dependent variables, as well as the relationship of the variables between the two levels.</p>
<p><b>Cited references to follow up on</b></p>	<p>Jakobsen, H.H., Blanda, E., Staehr, P.A., Højgård, J.K., Rayner, T.A., Pedersen, M.F., Jepsen, P.M., Hansen, B.W., 2015. Development of phytoplankton communities: implications of nutrient injections on phytoplankton composition, pH and ecosystem production. <i>J. Exp. Mar. Biol. Ecol.</i> 473, 81–89. <a href="https://doi.org/10.1016/j.jembe.2015.08.011">https://doi.org/10.1016/j.jembe.2015.08.011</a>.</p> <p>Jia, J., Gao, Y., Zhou, F., Shi, K., Johnes, P.J., Dungait, J.A.J., Ma, M., Lu, Y., 2020. Identifying the main drivers of change of phytoplankton community structure and gross primary productivity in a river-lake system. <i>J. Hydrol.</i> 583, 124633. <a href="https://doi.org/10.1016/j.jhydrol.2020.124633">https://doi.org/10.1016/j.jhydrol.2020.124633</a>.</p> <p>Zhang, W., Shen, J., Wang, J., 2021a. Linking pollution to biodiversity and ecosystem multifunctionality across benthic-pelagic habitats of a large eutrophic lake: a wholeecosystem perspective. <i>Environ. Pollut.</i> 285, 117501. <a href="https://doi.org/10.1016/j.envpol.2021.117501">https://doi.org/10.1016/j.envpol.2021.117501</a>.</p> <p>Zhang, X., Xie, P., Chen, F., Li, S., Qin, J., 2007. Driving forces shaping phytoplankton assemblages in two subtropical plateau lakes with contrasting trophic status. <i>Freshw. Biol.</i> 52, 1463–1475. <a href="https://doi.org/10.1111/j.1365-2427.2007.01776.x">https://doi.org/10.1111/j.1365-2427.2007.01776.x</a>.</p>
<p><b>Follow up Questions</b></p>	<ol style="list-style-type: none"> <li>1. How can the principles of hierarchical linear, multi-agent based, and vertically generalized production models be applied to network theory in order to model</li> <li>2. How does the type of body of water impact the environmental parameters that drive phytoplankton dynamics?</li> <li>3. What factors influence the extent to which broader-scale and smaller-scale environmental parameters connect to one another?</li> <li>4. What role does stratification play in influencing phytoplankton dynamics? How much impact do they have on other parameters?</li> </ol>

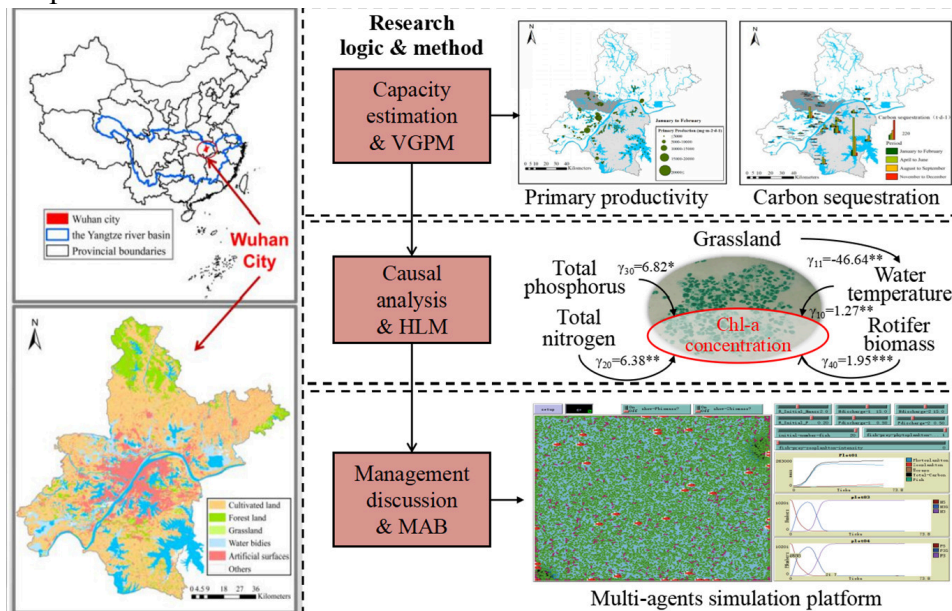
## NOTES

### Abstract:

- Understanding carbon sequestration capabilities of lakes is important in tackling climate change
- In order to understand the relationship between carbon sequestration and various environmental factors, specifically of a lake, an integrated method framework (see definition above, basically the bread and butter of your pro) was carried out.
- Important ramifications: offering additional lens on impact of climate change → important for policy makers.

- Three series of models:
  - Vertically generalized production model: Used for assessing carbon sequestration capacities (via chlorophyll a?), which was found to be prevalent in lakes
  - Hierarchical linear model: Used to identify the factors impacting phytoplankton dynamics
  - Multi-agents-based model: Used to identify possible mitigation and response strategies given changing carbon sequestration conditions
- Findings indicate that lakes have stronger sequestration capabilities, especially compared to ocean and forest ecosystems.
- Phosphorus, nitrogen, *Rotifer* biomass, water temperature, in that order, were driving factors impacted chlorophyll a (hereafter Chl-a) concentrations, which means it is the chemistry piece, going bottom-up, rather than the ecological piece (i.e. zooplankton, bottom-down) that appears to be impactful.
- Negative syn relationships between chl a and grasslands due to temperature
- Policy proposal based off the findings: Controlling the ratio of nitrogen to phosphorus and biomass of fish feeding on zooplankton in order to bolster C sequestration and maintain water quality.

### Graphical Abstract:



### Introduction:

- Oceanic carbon sequestration (hereafter C seq) is a major means through which mitigation of climate change can occur
- C seq depends on (1) the solubility pump (i.e.  $\text{CO}_2$  becoming more integrated into oceans due to increasing density in the air, and (2) the microbial pump, i.e. phytoplankton
- However, less research of lakes has been done than in oceans, so it is important to (1) understand their C seq capabilities, (2) understand factors influencing phytoplankton, and (3) suggesting action plans for policy makers.



- Moreover (similar to the findings that you have noticed for your own project, but a step even further almost), there is dearth in focus on identifying the factors that influence phytoplankton dynamics across multiple scales (i.e. not just biochemistry or nutrient stoichiometry alone, rather, environmental, ecological in conjunction with chemical, biological and physical. Different scale in the breadth of the parameters).
  - Much work has been done using PCA, redundancy analysis, canonical correspondence analysis, mainly on micro-physical, chemical and biological factors
    - (lists a whole bunch of studies which can be very useful for data...) light, suspended solids, turbidity/transparency, micronutrients, DO, pH, salinity, chemical oxygen demand (COD).
  - Focus has only been on these micro-level factors, but what about macro-level factors, like land use, pollution, etc.
- Using hierarchical linear model (HLM) is useful for separating impact of different IDVs of different scales on the DVs. It is good for analyzing the relationship phytoplankton has with any set of variables.
- Parameters establish the predictive model, which is important policymaking tool for enhancing C Seq.
- Need for complex adaptive systems to link different-scale parameters needed.
- Past device that have been used for policy recommendation use techniques like stock, flow, time delay, state variables and feedback loops to perform simulation.
  - There are limitations posed for this. These computational techniques fail to connect microbiological and macrological factors b/c intervariable interaction can only be weakly understood.
  - We want to understand the micro-chemical aspects of phytoplankton, and then extend the ramifications of that to C seq
  - Hard to describe phenomena that cannot be modeled by equations, e.g. random reproduction/predation
- However, Multi-agents based modelling (MAB) is a good solution to these limitations, as all variables are autonomous and interact with each other. With these conditions of interactivity, the impact of micro-scale conditions on macrological conditions can be observed
  - In this case, at the micro-level, for this study, phytoplankton, zooplankton, and other organisms, and macro-level, there was biomass, C seq, and water quality.
  - From this, a simulation was created, and it was able to model what would happen should recommended regulatory guidelines be followed.
- From this, >60 lakes in Wuhan, China were analyzed, as Wuhan, along w/ other Chinese cities have been making recommendations for lake management, so the ramifications of what they suggest ought to be evaluated.
  - Dealing w/ chronic eutrophication, biodiversity loss, water pollution
- Three-fold focus:
  - (1) Reveal C seq capabilities of lakes specifically
  - (2) Evaluate impact of environmental parameters impacting phytoplankton
  - (3) providing insights and policy recs for decision makers on climate change
- Three-fold hypothesis:

- (1) Lakes are capable of C seq
- (2) There exists relationships that are cross-sectional among phytoplankton, zooplankton, macro-level land cover, environmental parameters,
- (3) By linking the microbiological with the macrological, policymakers can be informed in decisions on management of lakes given climate change
- Crucial ramifications!

#### Materials and Methods:

- Wuhan contains a myriad of, >100 lakes, having large water supply.
- Pollution is rampant; 63 lakes. Poses threats to inhabitants drinking water and Yangtze, into which these water bodies drain
- 2018 site-monitoring data collected for these 63 lakes. Following data collected: name of lake, month & time of year, water depth and temp, clarity, pH, Chl-a concentration, Permanganate index, micronutrients, abundance of phytoplankton and zooplankton biomass. *Rotifera*, *Cladocera* and *Copepoda* were the three groups of zooplankton.

#### Overview of the methodology:

- Vertically generalized production model (VPGM) calculated primary productivity and C seq
- Nest data structure: used to establish relationship between aquatic and terrestrial parameters (novel)
- HLM: Determined factors impacting phytoplankton dynamics
- Principles of Lake-ecological-social management system (LESMS) was then applied to the findings to make policy recs.
- Multi-agent based modelling of the scenarios derived from this were performed on NetLogo.

#### VPGM:

- VPGM a type of vertical integrated model of clarity levels and common model of primary production. VPGM is initially used to calculate primary production in clear water. Following equation below was then used to account for the inland lake environments:

$$PP_{eu} = 0.66125P_{opt}^B \cdot \frac{E_0}{E_0 + 4.1} Z_{eu} \cdot C_{opt} \cdot D_{irr}$$

- variables include productivity in eutrophic zones, available light, light cycle (i.e. sunrise/sunset), Chl-a
- Other functions were used for calculating temperature: eutrophic depth, maximum photosynthesis rate, and another formula is used for water transparency.
- C seq = Avg net primary production \* lake surface area

#### HLM:

- C seq is dependent upon chl a. Here, we assume three levels of hierarchy impacting chl-a; one: micronutrients and water temperature from bottom-up, two: top-down impacts of the 3 zooplankton species, and land use, which accounts for pollution levels and water temp
- These factors impact C seq b/c they impact phytoplankton primary production and/or biomass
- Nested Structure Data was used to understand the impact of Chl a on marine ecosystems and organisms, and the change brought about by land use surrounding the lake, i.e. micro-marco variable relationship.

- HLM was what was used to analyze relationship between and within the micro-macro variables.
- Three steps to developing the HLM. unconditional means model (UMM), single variable random coefficient model (RCM) and the multivariate variables RCM and mixed effect model (MEM)
- UMM:
  - Tests for significance between the micro-macro variables.
    - $Level\_1 : Chl - a_{ij} = \beta_{0j} + r_{ij}$
    - $Level\_2 : \beta_{0j} = \gamma_{00} + u_{0j}$
    - $General Model : Chl - a_{ij} = \gamma_{00} + u_{0j} + r_{ij}$
  - From left to right: chlorophyll, beta (simplified out) is the mean level of individual chla concentration, the  $r_{ij}$  is the variation among individual data points among portion of a lake. Gamma is the average chla concentration of the entire lake, u is the variation between different lakes.
  - By analyzing if variances here are proportional (equal?), we get the following:
    - $Variance(Chl - a_{ij}) = Variance(r_{ij} + u_{0j}) = \sigma^2 + \tau$
    - Sigma looks at the partial lake level, “t” looks at the whole lake level. Then, they get:
      - $\rho_1 = ICC_1 = \frac{\sigma^2}{\sigma^2 + \tau}$
      - $\rho_2 = ICC_2 = \frac{\tau}{\sigma^2 + \tau}$
    - Bottom line: if  $ICC_2 < 0.059$ , Chla is independent from the lake level, and the groups have no difference, in which case there is no significance and only multi-regression is done, not HLM
- Single Variable RCM
  - RCMs used to assess how much impact level 1 variables impact the dependent variables, and if the level 1 coefficient and intercept of the linear model causes significant variation at level 2.
  - Level 1 variables of the study = water temp, micronutrients, 3 zooplankton species
- Mixed effect Model (MEM)
  - (see def above for the significance of it)
  - Relationships between water environment, aquatic organisms, and land use, and their impact of chl a are able to be understood now.
  - L2 = “contextual variable”; L1 = “Individual variable” in the equations

#### Multi-agent Based Modelling:

- MAB was used to simulate policy recs from the LESMS (which is above in the figure section, and about to be described below)
- Broadly, when doing the MAB simulation, we set up a conceptual model, define relevant parameters impacting the agents, build agents’ behavioural function, run and debug simulation, then experiment and predict scenarios, then analyze results

- Concept Model:
- From the LESMS, we have the five systems (see figure above): physicochemical factor environment subsystem (PES), zooplankton, phytoplankton and fish behaviour subsystems (ZBS, PBS, FBS), Policymaker Subsystems (PMS).
- Modelled the intra-inter relationship between all of these variables, minus PMS
- Biomass and debris of the behavioral systems comprises the C seq. N and P were used for PES.
- Transferred this info to the PMS.
- Biological manipulation and pollutant emission management were set up as parameters in order to help policymakers be able to meet goals of better water quality and C seq
- Used NetLogo. Each model of the cycle was counted as a tick, and within each of these cycles, the agents would interact and update their values given the rules set, in turn impacting all other individuals and overall environmental conditions

#### Results:

- (remember, all the computer model results are derived from the input of the 2018 data)
- VGPM used for calculating C seq (primary productivity and lake size as intermediaries of that process, ofc)
- For 2018, Average C seq was  $0.87 \text{ kg C/m}^2 \cdot \text{a}^{-1}$ ;  $0.56 * 10^6 \text{ tC} \cdot \text{a}^{-1}$  was the total C seq. April-June, Aug-Sep, Jan-Feb, and Nov-Dec, in that order, had the most C seq.
  - To my knowledge, a reference chl a.
- Larger surface area was positively related to C seq (make sense)
- Clearly, there is spatial and temporal heterogeneity in patterns of C seq, so it is important to identify those factors impacting this (surface area, what else...)
- It was first determined that it was appropriate to use HLM, given the process described above in the methodology, as the equations showed that the  $\text{ICC}_2 > 0.059$  was true.
  - This is the interclass correlation, meaning basically, that the threshold of some value has to be passed in order for there to be a significant noticeable relationship between the two levels of the hierarchy to do a HLM as opposed to only multi-regression analysis.
- First, single RCMs for the specific parameters were created for seeing which variables impacted the chl a levels the most significantly.
- (See analysis of table II above)
- Nitrogen, temperature, Phosphorus and *Rotifera* biomass were significant. N:P ratio and other two species were not. Indeed, *Rotifera* is known for its faster life cycles, small size, large population and big distribution, making it a big feeder on phytoplankton than the other 2 groups
- Isolating down from there, we get table 3 (see above). Using the gamma values, we know in this order: P, N, *Rotifera*, Temp; meaning the bottom-up chemical dynamics are more impactful than the top-down dynamics from predation.
- $\text{ICC}_1$ , the intra-class correlation, was significantly lower in the multivariate RCM, meaning these factors could be introduced into the MEM.
  - Why?
- Now, by implementing the level 2 parameters, the MEM was developed (impact of level 2 on chl a, plus the relationship with the significant level 1 parameters)

- Cultivated land being near the lakes with the phytoplankton had a positive correlation with the chl a, whereas there was a negative one for with the forest, mean chl a around cultivated lands was high, but low in forests. There was a negative correlation between grasslands and chl a by decreasing the water temperature (b/c  $\gamma_{11}$ ). Otherwise, no other results were significant. Land cover change did not impact N or P, nor the zooplankton
  - So little connection between the upper scale impacts and the lower scale impacts.
- Now the MAB:
  - Parameter values are entered, and then the program begins running, collecting and displaying the data.
  - Two scenarios of suggested management strategies were tested: changing point source pollution, and changing the fish culture dynamics.
  - C seq given point-source pollution:
    - Four experimental groups:
      - NAPA = 15 mg/L N, 0.5 mg/L P (of discharge)
      - NAPB = 15 mg/L N, 1.0 mg/L P
      - NBPA = 20 mg/L N, 0.5 mg/L P
      - NBPB = 20 mg/L N, 1.0 mg/L P
    - Temp was constant
    - These values of discharge come from the Chinese govt
    - (see analysis of results above)
    - Although pollution increased by a worrying degree b/c of more nitrogen and phosphorus, the phytoplankton and zooplankton were actually able to absorb it, without forming any blooms
  - C seq given changing fish cultures:
    - Four experiment groups:
      - P0Z0 = no fish (control)
      - P0Z1 = zooplankton-feeding fish (excl.)
      - P1Z0 = phytoplankton-feeding fish (excl.)
      - P1Z1 = omnivorous fish (excl.)
    - N & P (initial and discharge rates), and temp were constant
  - Not only was the P0Z1 best for C seq, but it was also good for absorption of N and P pollutants, therefore best for water quality.
    - P1Z0 = no reduction in pollution
    - P1Z1 = P pollution significantly reduced only
  - Meaning, a good recommendation would be to use more exclusively zooplankton-feeding fish

#### Discussion:

- Lakes have better C seq capabilities than oceans and forest ecosystems.
  - Data found here was greater than found at various points along China's coast. Previous studies show that ocean was 10x more efficient than forest
- Restates their findings on P, N, *Rotifer*, and water temp (ok lol)
  - But this does align with previous findings.
  - In "longstanding debate" about whether top-down ecology or bottom-up chemistry influences phytoplankton more, the data here indicate the former.

- With regards to cross-scale factors impacting each other, all they really found was that grasslands led to lower temperatures, which led to lower chl a at the partial-lake level.
  - Pretty disappointing, given what they purported. Or i guess, the fact of the matter is that smaller and larger scale parameters, under certain contexts, like this one, can be independent of each other.
- These parameters could become important metric by which policymakers choose to manage lake ecosystems.
- Given the results of our finding (and all of their significance/ramifications as discussed above), it is imperative that a better understanding between C seq and water quality is established.
- Policy proposal: Input N and P, as well as overall control over proportion, increasing biodiversity, purifying water (b/c pollution absorbed by phytoplankton), increasing biodiversity
- P0Z1 highest → Policy proposal: Input more zooplankton-feeding fish. With more phytoplankton there, there will be more primary production, absorption of nutrients, reducing pollution.
- However, caution should be present so as not to allow for blooms of phytoplankton to grow due to high biomass.
- Other suggests: Choose economically valuable zooplankton-feeding fish, and fix carbon, convert into biochar to meet carbon demands for the food supply.

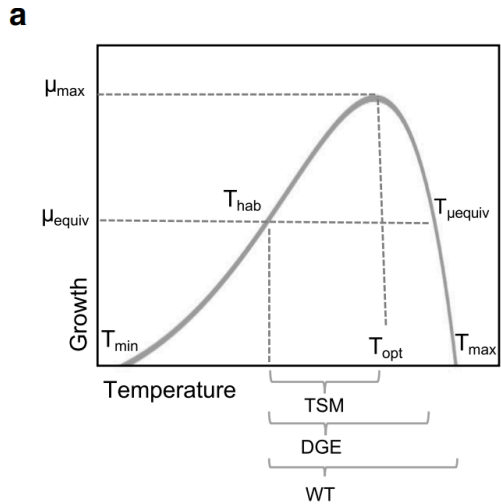
#### Conclusion:

- Summarizes article
- Point source pollution is positively correlated/matches correlation w/ c seq (essentially, the findings from NANB simulation and such)

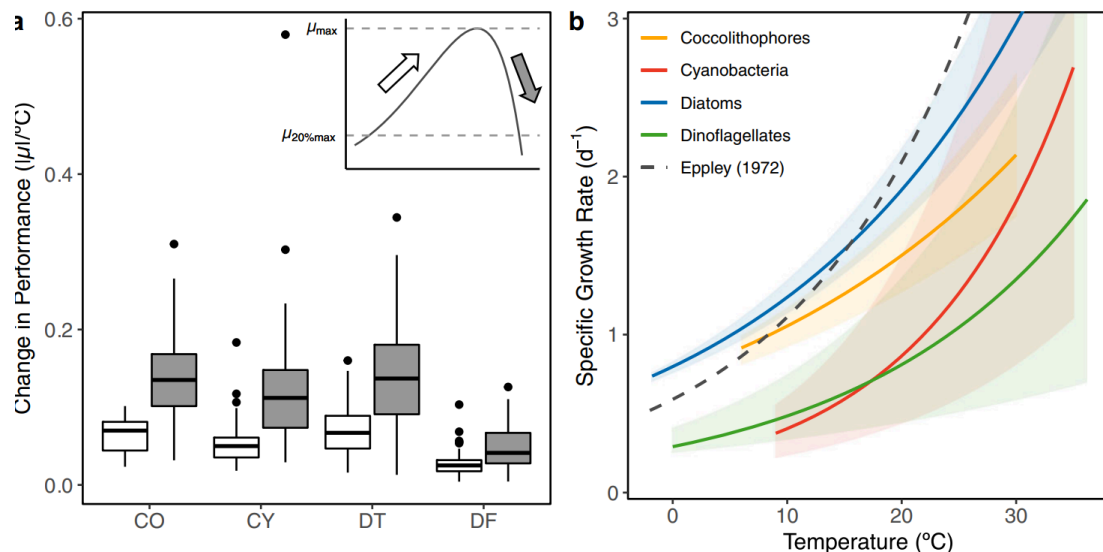
## Article #13 Notes: Marine phytoplankton functional types exhibit diverse responses to thermal change

<b>Source Title</b>	Marine phytoplankton functional types exhibit diverse responses to thermal change
<b>Source citation (APA Format)</b>	Anderson, S. I., Barton, A. D., Clayton, S., Dutkiewicz, S., & Rynearson, T. A. (2021). Marine phytoplankton functional types exhibit diverse responses to thermal change. <i>Nature Communications</i> , 12(1). <a href="https://doi.org/10.1038/s41467-021-26651-8">https://doi.org/10.1038/s41467-021-26651-8</a>
<b>Original URL</b>	<a href="https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8571312/">https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8571312/</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	The article does not provide any keywords.
<b>#Tags</b>	<a href="#">Definition of Four Major Groups of Phytoplankton</a> <a href="#">Alternative General Data Analysis Software to Sheets or Excel</a> <a href="#">Basis for Thermal Equation and Criteria Used in Study</a>
<b>Summary of key points + notes (include methodology)</b>	Phytoplankton phylogeny is known to be heavily diverse, however, the diversity of traits among the different groups therein, and in turn, the environmental and climatic impact, is not well understand. Thus, this study focuses on determining any difference between thermal norms among four major functional groups: dinoflagellates, diatoms, cyanobacteria, and coccolithophores. It seeks to compare temperature with growth rate and geographic distribution among these four groups. Data on multiple strains' thermal properties, including their optimal, maximum, minimum and habitat temperature, and various measures derived from the differences between these quantities was compiled. It was found that the groups significantly differed from one another in terms of their thermal-sensitivity with growth rate, diverging from the commonly used Eppley formula used to measure this relationship. In performing analysis using Kruskal-Wallace and Dunn Tests, it was found that the growth rate had a positive exponential relationship with temperature, with diatoms being projected to have the greatest increase in growth rate. Geographically, the common trend of poleward migration is reinforced, with all four groups' abundances projected to decrease in the equator, but increase in the middle and polar latitudes. Future steps include modelling the impact of these changing migration patterns on the food web and climate more directly, as well as enhancing the thermal models developed in this study.
<b>Research Question/Problem/ Need</b>	How do thermal norms among different phytoplankton groups vary, what ramifications does that given ocean warming, and how does this affect migration?

## Important Figures



This figure (3a) provides a template model for the growth rate curve given temperature among the different groups.  $\mu_{\max}$  is the peak growth rate, which occurs at the optimal temperature  $T_{\text{opt}}$ . However, there exists some range of temperatures in the habitat under which a given minimum  $\mu$  can exist (for example,  $\mu$  has minimum value of 1 between 30 and 33 degrees, with minimum value being lower for larger range of temperatures), ranging from the habitat temperature,  $T_{\text{hab}}$ , and the temperature on the other end of the thermal-growth curve with the same  $\mu$ ,  $T_{\text{equiv}}$ . More broadly, there exists a  $T_{\text{max}}$  and  $T_{\text{min}}$  within which a functional group is viable. TSM, the thermal safety margin, is  $|T_{\text{hab}} - T_{\text{opt}}|$ , the temperature range within which the phytoplankton can viably grow without strain. The warming tolerance, by contrast, is  $|T_{\text{max}} - T_{\text{opt}}|$ , which represents functional group growth in any capacity. The DGE, the distance to the growth equivalent, is the range between  $T_{\text{hab}}$  and  $T_{\text{equiv}}$ .

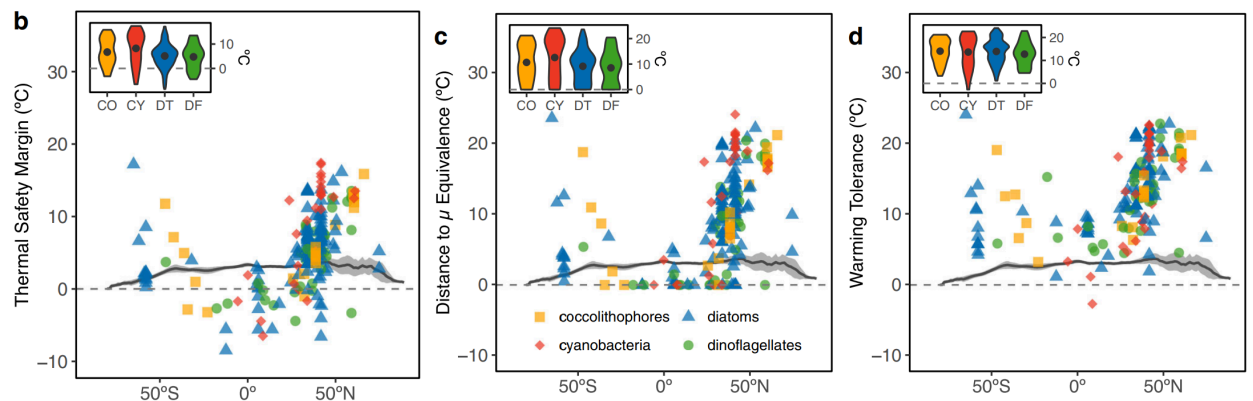


This is figure 2 of the study. The left diagram contains a series of boxplots that measure the change in growth rate, given the temperature. Unshaded boxplots are for when the temperature ascended towards optimal temperature, and the shaded boxplots for when the temperature was past it (but still within the warming tolerance/viability range). Change appears to be most restricted for the dinoflagellates, followed by the cyanobacteria, coccolithophores, and diatoms.



The median change in performance is about  $0.03 \mu/\text{C}$ , with a tight, symmetric range between about 0.02 and 0.05 approaching  $T_{\text{opt}}$ . Cyan., Cocco., and Diat. have medians of about 0.04, 0.05 and 0.05 respectively at this thermal stage, and all of their distributions are more variable, being skewed to higher changes in performance. These three groups have at least 50 to 75% of the distribution in performance changes above the entire distribution for dinoflagellates. Meanwhile, a similar pattern is discernible when approaching  $T_{\text{max}}$ , past  $T_{\text{opt}}$ , the only difference is that all distributions are more varied (but with fewer outliers), and the median performance change is higher.

Meanwhile, the figure on the right illustrates the various diversions of each functional group's growth rate curve (given temperature) from the standard Eppley Curve. Immediately, a few key characteristics can be discerned. First, there is the viable temperature ranges. Cocco group has the most restricted temperature range, with a lower bound of about  $6-8^{\circ}\text{C}$ , and upper bound of just under  $30^{\circ}\text{C}$ . Cyanobacteria cannot survive under  $9^{\circ}\text{C}$  (as discussed below). As discussed below, the Cyans have a higher exponent than the Eppley Curve, whereas the other three groups are smaller. That is, as temperature increases, this group's growth rate increases at a faster rate. This is not to be confused the growth rate itself. In that arena, Diats are the winner, having the highest  $\mu_{\text{max}}$  at all temperatures. They also have the smallest standard errors, as indicated by the shaded regions (which represent 95% confidence intervals, the Markov Chain bootstrapping), which is likely attributable to its large sample size. The amount of spread increases, with Diats being the most finely distributed, followed by Cocco, Dino, and Cyan groups. This is because the variability in thermal growth rates among individual strains gradually increases, causing this higher variation.



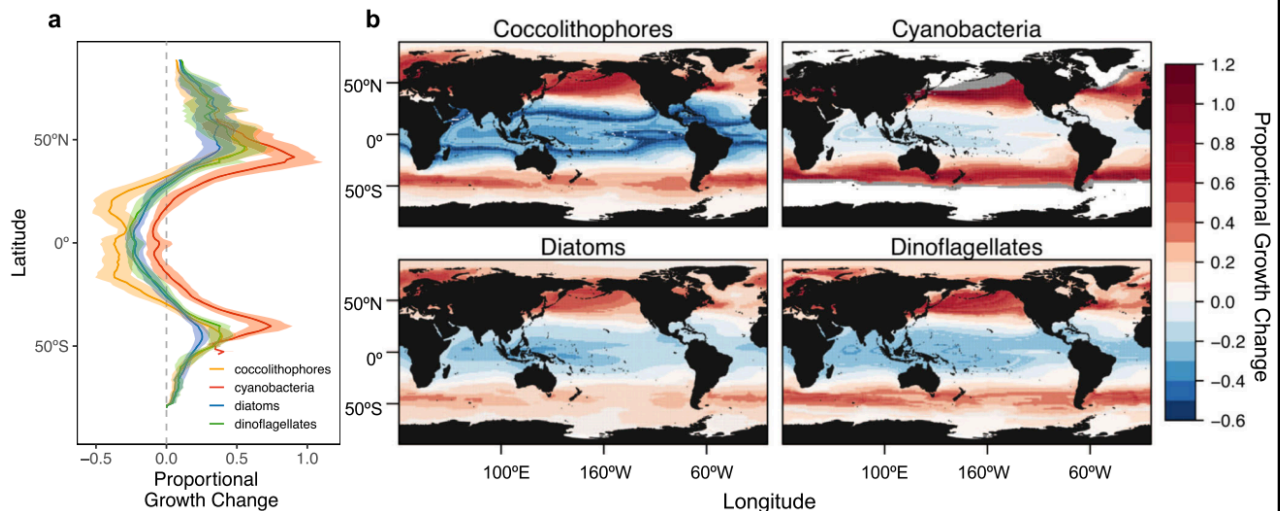
From left to right, figures 3b depict the thermal safety margin (TSM) of the phytoplankton groups. This is measured by  $T_{\text{hab}} - T_{\text{opt}}$ , and represents the ability for performance to be maintained without strain incurrence. The gray line indicates expected increase in SST for 2100, with the shaded region indicating longitudinal variation. Figure 3c depicts DGE, which represents the entire thermal range under which  $\mu_{\text{max}}$  (or greater) is maintained, which accounts for temperatures both above and below the optimum. Finally, figure 3d measure warming tolerance, the entire thermal range under which the given group can even survive. For all three figures, individual strain results are plotted against latitude.

Starting with 3b, the datapoints seem to indicate that cyanobacteria are only found in the northern hemisphere (or, at the very least, the ones sampled were all from this range only). At the equator, the temperatures presently appear too be below the optimal level, given that TSM is negative,

and for  $T_{\text{hab}} - T_{\text{max}}$ ,  $T_{\text{hab}} < T_{\text{max}}$  would have to be true. Using a similar logic, at the middle latitudes, the  $T_{\text{hab}}$  is above the  $T_{\text{opt}}$ . Measuring correlation between latitude and thermal safety margin, while stratifying for each of the four functional groups, there seems to be a trend of upward concavity between these two factors. In context, that means the mid-latitude areas are warmer than the optimal temperature, whereas equatorial ones are cooler, conforming with the above observations. Once more, this is not the case for the cyanobacteria, which were not sampled from southern hemisphere.

For figure 3c and 3d, the same geographical patterns apply. Looking closer at 3c, there are no values below 0, which, numerically, makes sense, as the distance between habitat temperature and the equivalent growth rate achieved at the other end of the thermal spectrum, is an exclusively positive measurement. This may be due to sampling (which has been a trend throughout all studies—the dearth of data from the Southern hemisphere), but it would seem as though the thermal equivalent allows for a much broader range of temperatures for a given growth rate in the northern latitudes. However, a similar parabolic pattern follows from figure 3b. This suggests that for equatorial regions, the thermal allowance is more restricted, as a small distance between the  $t_{\text{han}}$  and the  $t_{\text{equive}}$  would be much more condensed. This could be in part due to the more cooler (relative to  $t_{\text{opt}}$ ) temperatures, thermal homogeneity, or some other factor.

For figure 3d, the same patterns in spatial abundance of the functional groups, as well as the parabolic relationship, still hold true. The more equatorial a latitude, the lower the warming tolerance, which, given that this area of the ocean is already the warmest, make sense.



Both figures 4a and 4b come together to form a narrative of the geographical trends predicted by the modeling carried out in this study. On the left, figure 4a illustrates a relationship similar to figure 3. Whereas the equatorial phytoplankton are expected to decline in terms of abundance, phytoplankton populations are projected to increase in the middle latitudes. The lines represent the mean proportional change prediction, with the shading representing deviation. Analyzing the troughs and peaks of each group, it seems that the coccolithophores are poised to experience the greatest loss, while cyanobacteria the greatest growth. Figure 4b is a visualization of 4a, providing a global map that is shaded, with cool colors indicating decline and warmer ones growth. In all four cases, with the warm colors in the polar/midlatitude regions, and the cool colors in the equator, the findings from figure 4a are reinforced. This is supportive of the overall trend of

poleward migration that has been noted in other studies.

**VOCAB:  
(w/definition)**

**Coccolithophores** - A major group of phytoplankton that inhabits the ocean, particularly at the shallower layers including greater amounts of sunlight. They are known for inhibiting carbon sequestration mechanisms via the production of  $\text{CaCO}_3$ . They have a ring-shaped structure, and are coated with  $\text{CaCO}_3$ .

**Cyanobacteria** - A highly diverse group of prokaryotic and autotrophic gram-negative bacteria known for being among the first organisms to produce oxygen. Thus, they are major climate regulators.

**Dinoflagellates** - A group of phytoplankton known for their unique and complex cytological structure, most notably their two flagella.

**Diatoms** - A major group of phytoplankton, comprised of algae and microalgae. This group alone has been estimated to be responsible for 20 to 50% of production of atmospheric oxygen annually.

**Diazotrophs** - Marine bacteria that absorb and convert atmospheric nitrogen into ammonia and other nitrogenous nutrients usable in an oceanic environment.

**Eppley Curve** - The exponential curve created by Richard W. Eppley to model the impact of SST on phytoplankton growth via the following equation:

$$\mu = a^{b \cdot T}$$

Where  $\mu$  is the maximum growth rate of a given phytoplankton species,  $T$  is the temperature.  $a$  and  $b$  are parameters that are constantly changing as more data using quantile regression (explained below) is received.  $b$  is often referred to as the Eppley exponent, and it measures the degree of growth the maximum growth rate is increased due to temperature, whereas  $a$  is the growth rate at  $0^\circ\text{C}$ .

**Static Heat Capacity** - The amount of heat (in thermal energy- the speed and collision of the molecules) exposure given to an object in order to increase its temperature (heat is the energy, temperature is the measure).

**R-strategists** - Species that have fast reproduction rates and life cycles, meaning their populations evolve very quickly (very applicable when considering bacteria and other organisms).

**Ectotherm** - An organism that relies on external sources of heat (as opposed to an endotherm, which does that opposite).

**Quantile Regression** - An extension of linear regression where medians, or other quantiles of data (that is, portions of the bell curve's probability density function - broad term applicable to quartiles, deciles, percentiles, etc.) are used to form a regression model between two or more variables (there are multi-variate versions). Using other measures of central tendency and variation can be insightful in revealing trends between variables.

**Akaike information criterion with correction (AICc)** - A critical tool for assessing the quality of statistical models, and choosing the best options. The amount of information omitted from a model is measured with this tool, with a cost-benefit analysis balancing the degree of fit with information loss to evaluate the model.

**Markov chain marginal bootstrap** - A special type of bootstrapping technique. Bootstrapping is essentially the development of a confidence interval for resampled data, where the population parameter values are based off the results from the original sample, rather than hypotheses for the population at large. The Markov chain marginal bootstrap in particular focuses on eliminating computational cost by converting multi-dimensional algorithms into single-parameter equations.

	<p><b>Kruskal–Wallis Test</b> - The nonparametric equivalent of a one-way ANOVA test. Typically in one-way ANOVA, there is an assumption that all of the population means are equivalent. However, in Kruskal-Wallis, no such assumption is made. This is because the focus in this test is on categorical data placed under a ranking system. Consequently, a chi-squared statistic is used rather than a F-statistic.</p> <p><b>Dunn’s Multiple Comparison</b> - Just as Kruskal-Wallis is the nonparametric ANOVA using categorical data/chi square statistic, the Dunn’s Multiple Comparison test is analogous to the Post-HOC Tukey tests, doing pairwise comparisons to note the specific locations of any statistically significant differences.</p> <p><b>Norberg Curve</b> - A sort of specialized function/curve that is used to model phytoplankton thermal relationships and their statistical/empirical values.</p> <p><b>Pearson’s Correlation</b> - The formal statistical name behind “r”, the correlation coefficient.</p>
<p><b>Cited references to follow up on</b></p>	<p>Bopp, L. et al. Multiple stressors of ocean ecosystems in the 21st century: Projections with CMIP5 models. <i>Biogeosciences</i> 10, 6225–6245 (2013)</p> <p>Allen, A. P., Gillooly, J. F., Savage, V. M. &amp; Brown, J. H. Kinetic effects of temperature on rates of genetic divergence and speciation. <i>Proc. Natl Acad. Sci. USA</i> 103, 9130–9135 (2006)</p> <p>Steinacher, M. et al. Projected 21st century decrease in marine productivity: a multi-model analysis. <i>Biogeosciences Discuss.</i> 7, 979–1005 (2010).</p> <p>Ward, B. A., Cael, B. B., Collins, S. &amp; Robert Young, C. Selective constraints on global plankton dispersal. <i>Proc. Natl Acad. Sci. USA</i> 118, 1–7 (2021).</p> <p><i>Citations mentioned in the methodology that are relevant to understanding the question, not necessarily useful for the project in the long-term, however:</i></p> <p>Thomas, M. K., Kremer, C. T., Klausmeier, C. A. &amp; Litchman, E. A global pattern of thermal adaptation in marine phytoplankton. <i>Science</i> 338, 1085–1088 (2012).</p> <p>Eppley, R. W. Temperature and phytoplankton growth in the sea. <i>Fish. Bull.</i> 70, 1063–1085 (1972).</p> <p>Bissinger, J. E., Montagnes, D. J. S., Sharples, J. &amp; Atkinson, D. Predicting marine phytoplankton maximum growth rates from temperature: Improving on the Eppley curve using quantile regression. <i>Limnol. Oceanogr.</i> 53, 487–493 (2008).</p> <p>Rynearson, T. A. Estimated thermal capacities for phytoplankton strains. Biological and Chemical Oceanography Data Management Office (BCO-DMO), <a href="https://doi.org/10.26008/1912/bco-dmo.839713.1">https://doi.org/10.26008/1912/bco-dmo.839713.1</a> (2021).</p> <p>Kremer, C. T., Thomas, M. K. &amp; Litchman, E. Temperature- and size-scaling of phytoplankton population growth rates: Reconciling the Eppley curve and the metabolic theory of ecology. <i>Limnol. Oceanogr.</i> 62, 1658–1670 (2017).</p>
<p><b>Follow up Questions</b></p>	<ol style="list-style-type: none"> <li>1. How are ocean temperature, phytoplankton metabolic capabilities, as well as migration intertwined with another, and how do their changes impact the trophic pyramid?</li> <li>2. What physiochemical relationships exist between micronutrients and temperatures, and how does that impact phytoplankton populations?</li> <li>3. How do differing phytoplankton thermal norms impact the taxonomic composition of varying strata, and how does that impact the transfer of light resources?</li> <li>4. How can a continuous time series of the thermal trends depicted in this and similar studies be developed?</li> </ol>

## NOTES

## Abstract:

- Phytoplankton are phylogenetically diverse, and have different attributes when it comes to thermal conditions
- Diatoms, Coccolithophores, dinoflagellates, and cyanobacteria, four major components of primary productivity and ecological biomass/organic matter.
- Each of their thermal growth properties are different, and their equations derived
- Impact on growth rate and geographic distribution of these four major groups were tested using models of temperatures from 1950-1970 (past/given data), and the 2080-2100 (future/projected data).
- It is found that growth maxima thermal dependencies, thermal ranges and temperature coefficients all vary for each of the four groups, indicating diverse responses, and thereby changes in taxonomic composition and distribution (which in turn has major ecological/climatic impacts).
  - Contrasts with previous findings from the Eppley formula.
- Therefore, it is predicted that there will be significant changes to phytoplankton tax comp
- Specific findings include fewer lower-latitude coccolithophores and greater amounts of cyanobacteria.

## Introduction:

- Phytoplankton sequester 45 gigatons of CO<sub>2</sub>, vulnerable to climate change.
- However, understanding the impacts different groups face has hitherto been challenging due to diverse phylogenetic/evolutionary background.
- Whether primary production increases or decreases in phytoplankton depends on temp, and the biodiversity of the habitat ranges of this parameter that lie therein
- Presently, the same thermal capacities are applied to all groups of phytoplankton, making estimations too broad.
- The ramifications of different thermal capabilities in different species has yet to be explored.
- These varying capabilities shall result in different migratory responses and abundance changes among different phytoplankton species, modifying tax comp
- One major ramification of changing geographic ranges is that nutrient cycling, and other biogeochemical cycles are to be highly altered.
- Hence, this study defines the different thermal capabilities found among species, determining the impact on growth, distribution, and other dynamics.
- Dinoflagellates, coccolithophores, cyanobacteria and diatoms, 4 major functional groups, assessed. Distribution and abundance measured by using past data and future projections of temperature from Earth System Models.
- While there are other parameters that impact phytoplankton dynamics, temperature is a stronger indicator compared to pH, nutrient concentrations, light, etc. b/c they depend on complex biological processes, and have less confidence in terms of model accuracy/prediction. Temperature is a strong indicator of phytoplankton growth and abundance.
- Findings validate the hypothesis (different temp coefficients, growth maxima thermal dependencies, and thermal ranges causing different trends in phytoplankton)

## Methodology:

- Growth Data Compilation:
  - Thermal growth rates of the four functional groups assessed (w/ sample sizes given. Unfortunately, they are not equal (but maybe the next best step would be to be proportional? Idk... )
    - 200 coccos, 500 cyans, 1800 diats, 750 dinos.
  - Built off pre-2012 data for thermal growth rates from previous author, Thomas et al., using his same selection criteria (something that ought to be enumerated), though with modifications:
    - Lowered minimum rate to 80  $\mu\text{mol}$  of photons meters squared per second. (from 100)  $\rightarrow$  allowing for more representation of cyanobacteria (so there is a disparity in data availability here... major factor to consider?)
    - No data attained from experiments where nutrient cocentrations were studied (possible sources of confounding)
    - Diazotrophic species of cyanobacteria were not considered due to their physiological differences, resulting that may add confounding variable to the changing dynamics.
      - Separate analysis was performed. However, the data size was too small, and so no data was reported- future area of focus!
    - Dinosaurs only worked with in the lab, possibly could have ingested bacteria
    - Data analysis using GraphClick
    - 243 strains, >3k thermal growth rates measured.
    - Thermal reaction norms were instrumental in producing predictions on how the phytoplankton would react. (Equations used from Thomas et. al)
    - For the new post-2012 strains that were added in, the thermal attributes were estimated using the “maximum likelihood approach” from Thomas et al. 2018, and R (the programming language  $\rightarrow$  reactive programming)
- Climate Data:
  - Focus here was on the temperature increasing, so it easily follows that temp data had to be extracted from climate databases  $\rightarrow$  sea surface temp (SST) specifically
  - Coupled Model Intercomparison Project phase 5 (CMIP5) model projections
  - <http://climexp.knmi.nl>
  - SST warming was estimated based on the existing 1950-1970 data as a baseline, and, under the worst-case greenhouse has scenario, produced results for 2080-2100.
  - Done on a latitudinal basis (each had their own sd and x-bar)
  - $T_{\text{hab}}$  = habitate temperature for the phytoplankton strain. Attained via comparison w/ mean data from ‘50-‘70.
- Thermal Dependencies:
  - $\mu_{\text{max}}$  = maximum growth rate with temperature; expressed w/ exponential function
  - Key citations here (I wish they would not do this): Eppley was 1st to describe this function. Bissinger et al. later standardized methodology through which curve-fitting occurs
  - Bissinger followed via fitting a 99th quantile regression model to logarithmically transformed data on growth rates for each of the groups (under warming conditions, using quantreg package from R)

- 99th quantile was used, although it requires a lot of data and the results are vulnerable to many sensitivities, however, this method is superior:
  - Compared to a plain LSRL and the metabolic theory of ecology, the quantile regression is more comprehensive; LSRL only includes data below thermal optima, and the metabolic theory only focuses on  $\mu_{\max}$  w/o regard between the direct relationship with temperature.
  - LSRL fits to the 50th percentile data, which is undesirable in this context: the focus is on the *maximum* growth rates, hence we want the 99th percentile, or, to be more general, the 99th quantile of the data.
  - Quantile regression also does a better job of illustrating the dispersal of growth rates and the variable responses to temperature than LSRL
  - Another reason quantile regression is superior to metabolic theory is that there are no assumptions needed regarding cell size, which varies widely among different species.
- Phytoplankton growth rates and  $\mu_{\max}$  are statistically significant among the different functional groups. Thus, each quantile regression (applied to the 4 functional groups), was applied to these parameters separately.
- AICc (defined above) was used to make the model even stronger.
- Hence we have the following equation:
  - $$\mu_{\max}(T) = a \cdot e^{b \cdot T}$$
  - Where  $\mu_{\max}$  = maximum growth rate, given temperature (the function of T, temperature) - definition offered above, but to reiterate (only difference is that the base is e, rather than a):
  - a is the parameter representing the maximum growth rate at 0 °C, while b is the rate at which the maximum growth rate increases as the temperature does (meaning there is a positively linear, or at least positive form of relationship between the two)
  - 95% confidence interval was created for the curves of each of the functional groups via the use of 10k iterations of a Markov chain Monte Carlo bootstrap. This is ideal as this produces a computational algorithm that calculates only a one-dimensional equation given multiple parameters, that is, multiple dimensions.
  - From these exponential functions, the temperature coefficient  $Q_{10}$ , a measure of the rate at which  $\mu_{\max}$  increases for every increase in temperature by 10°C, was attained, allowing for the assessment on metabolism.
  - This departs from previous studies, where the functional groups had the same b value, and thus the same  $\mu_{\max}$ .
  - Activation energy was then calculated based off the exponential model/ $Q_{10}$
- Static Thermal Capacity and Statistical Analyses
  - Thermal safety margin (TSM) and warming tolerance (WT) were used as metrics to calculate thermal capacity (see above definition).
  - These metrics account for the thermal attributes of the organism, given habitat temperature, and are used to calculate the temperature at which performance begins to decline (TSM) and then when viability is lost (WT). The habitat is assumed to be the temperature at one point, hence the label “static”.

- To measure these metrics, each strain's thermal optimum and maxima was calculated:  $TSM = T_{opt} - T_{hab}$ ;  $WT = T_{max} - T_{hab}$
- $T_{max}$ : Dino = 38, Dia = 115, Cyan = 31, and Cocco = 24 °C
- However, it has been found that phytoplankton do not operate at their maximum growth rate, even at temperatures below the optimal one. Therefore, (and I judge this to be a decision to be a bit dubious), the authors presume that the same holds true for temperatures above the optimal level (I suppose this does imply the presence of some gradient, so maybe this assumption is not totally fallacious).
- As a mathematical consequence, a temperature between  $T_{max}$  and  $T_{opt}$  was assigned as  $T_{\mu_{equiv}}$ , this temperature being the maximum temperature at which the growth rate of the phytoplankton remains the same as the habitat. The spread of temperature covered between the  $T_{hab}$  and  $T_{\mu_{equiv}}$  was coined distance to the growth equivalent (DGE), that is,  $T_{\mu_{equiv}} - T_{hab}$ .
- Variation in these thermal attributes were then measured using Kruskal–Wallis tests, followed by the Dunn's multiple comparison tests at  $\alpha = 0.05$
- Future Metabolic Projections
  - The proportional growth change  $(\mu_{future} - \mu_{past})/\mu_{past}$  for each strain among the four functional groups was calculated as a first step to understanding what impact the change in temperature had on phytoplankton growth rates and those ramifications.
  - This relationship was modeled for each global grid cell (at 1.25° resolution)
  - Viability was defined as  $\mu_{20\%max}$ , which, as the name suggests, is 20% of the maximum growth rate. 20% was determined as the percentage, given that authors noted that species presence declined when starting off at lower percentages. However, 80% is typically used. (Clear reason for choosing 20% rather than 80% does not really make sense)
  - The growth change was then collected among all strains, and for each genus, the median value was used. Since the distribution is not normal (see figure 3a and other functions above), this is a better summary statistic (hence the quantile regression as well).
  - The viability criteria was applied at all latitudes to also assess whether there would be cyanobacteria, as this group is expected to grow greatly in range.
  - Spatial trends were enumerated via the average population growth rate change across different latitudes.

#### Results and Discussion:

- Disparities in Thermal Response
  - (Again, a broad range of temperatures from a wide area was used)
  - Through the data processing methods employed, authors claim that the data is 72% different from “typical data” (interesting, possibly controversial claim...)
  - Growth rate vs. time function fitted to a Norberg curve. Norberg curve was also utilized to find the growth rate maxima among individual strains.
  - Again, cutoff of 20% of the  $\mu_{max}$  was used, eliminating bias (skew) in the data brought about due to ambiguous thermal minimum.
  - The absolute value of the difference of the thermal reaction norm slope from  $\mu_{max}$  to 20% of  $\mu_{max}$  was taken, and placed over the temperature in Celsius. Meaning, the slope was  $|\mu|/^{\circ}C$ . Intuitively this makes sense, as the growth rate is being



- altered by the changing temperature, so the performance change would be some measure of growth rate changing, given the temperature (independent variable).
- This slope was also used in understanding the skew of the data (the slopes of the growth rates given the temperature changes)- the greater degree of positive difference from  $T_{opt}$  indicated that these data were skewed to lower values (which makes sense, b/c if the difference exceeds  $T_{opt}$ , the values would have to be really low)
  - The article first cites numerous data and evidence to suggest that the thermal attributes and responses among the four groups are not homogenous.
    - Cyanobacteria and Coccolithophores have notably limited temperature range:  $<9^{\circ}\text{C}$  and  $>30^{\circ}\text{C}$  respectively, neither can survive. Meanwhile, diatoms have a wider thermal niche.
    - (Somewhat obviously), the geographic distribution depended upon the thermal gradient
    - No significant correlation with the sample size, or the absolute isolation latitude, according the Pearson's Correlation test.
    - (see analysis of Figure 2 above)... the small changes observed in the Dino. group resulted in flatter curves. These trends (which match previous results, as this case with most results, unless otherwise stated), may be attributable to the daily vertical migration that this group engages in. This would have led to a lower amount/variability in growth rate, in exchange for greater thermal breadth.
  - The article then provides data/discussion for  $Q_{10}$ , the growth rate change experienced for every increase in temperature by  $10^{\circ}\text{C}$ , and Epply curve:
    - Temperature vs. growth rate maxima modeled via exponential function, in conformity with most models of Earth systems, primary production and growth rates.
    - Again, the main purpose of this article is to highlight the different thermal norms in the functional groups. Hence, the  $Q_{10}$  is proven to be statistically significantly different among the functional groups, diverging from methodologies of the previous studies (Kruskal-Wallis Test used).
    - 99th Quantile Regression Utilized (advantages of this choice described above in the methodology section), but main thing to note here is that this method allowed for the exponent to numerically characterize the relationship between group growth rate and temperature, allowing for  $Q_{10}$  to be calculated.
    - Epply curve characterizes the impact of temperature on growth rates as 1.88 (this is the b constant- measures the degree of increase the maximum growth rate experiences due to temperature- sorta basically just  $Q_{10}$ ).
    - However, the functional groups differ from this greatly (even when evaluated as one collective entity), illustrating the sheer thermal variability observable among different phytoplankton groups. Collectively,  $Q_{10} = 1.46$ 
      - Indeed, literature has been reaching a consensus that Epply Curve (1972) is an overestimate
    - Exponent values are as follows for each of the groups:

- Coccus (1.42), Diats (1.55), Dinos (1.67), and Cyan (2.13)
- Y-ints (i.e. growth rate at 0°C), were significantly lower for Cyan and Dinos (however, note that Cyan can't survive under 9°C, anyway)
- Cyan > other groups → significant
  - Contrary to the finding explained in the previous bullet at this level, authors argue that Eppley is not merely an overestimate, rather, its error depends upon the  $Q_{10}$  of the given group
  - However, there is another lens of analysis to consider: comparing the  $\mu_{\max}$  of the functional groups at a given temperature.
  - Despite having the smallest exponents, at 20°C, Diatoms and Coccus had the greatest growth rate (based off model predictions) → could indicate a superior ability to compete, as, despite a low temperature-sensitive growth rate change, they still have a higher growth rate at this given temperature, b/c they can, well, compete better, thus allowing for ^growth
  - Diatom trends are particularly notable, as they have the highest predicted growth rate at all temperatures compared to the other groups (see analysis of figure two above).
    - Indicative of them being r-strategists, groups that maximize the growth rate when under favorable conditions. Also indicates that this group would survive well under nutrient-sparse conditions, consistent with the findings of studies that analyze cocco-diat competition in oligotrophic systems.
    - Indicative of Diat's strong ability to adapt to unideal thermal conditions (excessively warm conditions, specifically).
  - All of these dissimilar thermal-sensitive growth rate trends among the four groups speaks to their unique evolutionary history (> 1b years), which included 2 instances of endosymbiosis
- Thermal Capacity Across Latitudes
  - The article re-explains how it defined DGE, Thermal Safety, and Warming tolerance. However, key thing to note is that DGE is special for taking into account the growth rate staying equivalent at temperatures exceeding  $T_{\text{opt}}$ , is useful, given that temperatures are warming. Overall, a more full picture is offered.
  - (see figure 3 analysis)
  - As indicated by figure 3, even though many of the mid-latitudes had temperature levels above  $T_{\text{opt}}$ , death was prevented through the large warming tolerances, something seen across all functional groups. However, this indicates that there was a significant amount of strain placed on performance.
  - (see above, but good to reiterate) Overall finding in mid-latitudes: there are temperatures that are above optimal moreso in this region, inhibiting performance though not causing death, with the warming tolerance of the functional groups still buffered from that. Also, the DGE is able to indicate that growth rate itself is not too strongly impacted either.
  - Explanation offered for the parabolic trends noted in figures 3b-3d:

- Although the equatorial phytoplankton have higher optimal and maximum temperatures, the ocean habitat temperature here does not increase. (meaning homogenous temperature across the ocean (?))
- This may indicate that the equatorial phytoplankton may struggle to adapt to the warmer temperatures. (Well, this could also just be math; the same thermal norms remain, but since the  $t_{hab}$  is already warm, the difference will start off lower. However, maybe a relationship analogous to the mid-latitudes will emerge, only that the  $t_{bab}$ ,  $t_{opt}$ , and all of those other parameters are all higher.
- Parabolic trend has been observed in other species as well.
- There was no significant difference between the PFTs with regards to warming tolerance and DGE, but cyanobacteria were significantly different w/ TSM compared to dino and diats, specifically indicating that they are more likely to inhabit abnormally cooler temperatures. (Makes sense — they prefer higher temps)
- Changing Rates and Shifting Ranges
  - Model for the ends of “simulating” changing geographic dist *does* take into account temperature gradients, migratory capabilities, and phenotypic variation w/n groups
  - Rather than actually simulate the movement of the phytoplankton, it was assumed that any region containing the proper temperature for phytoplankton to thrive could contain them (sort of a spatial probability approach).
    - Use of 20% of the maximum growth rate (methodology)
    - Use of CMIP5 models for SST + comparison between 1950-1970 and 2080-2100, calculating the median proportion of growth rate at each latitude
  - Differing changes in phytoplankton abundance = different composition, possible changes in competitive attributes
  - Cyanobacteria can now inhabit regions in that were once too cold for them due to ocean warming (e.g., Gulf of Alaska, Norwegian Sea)
    - Imposition of competition in new areas + similar patterns in the cocco
  - Coccolithophore decline in equator leads to less calcium production and chemical interaction, altering alkalinity.
  - DGE exceeded the thermal capacities of most strains at the equator, so equatorial decline does makes sense.
  - Opposite trend in mid/polar latitudes: especially in Southern ocean, where iron concentration due to ice melt is expected to increase.
  - But this increase might be inhibited by nutrient availability, particularly the nitrogen deficiencies expected to arise in the ocean.
  - Model accounts for temperature alone, not considering other factors that impact growth rate and migration.
  - Overall, there multifarious impacts that warming ocean temperatures are expected to have on phytoplankton. This lack of homogeneity is in line with other studies’ findings.
  - Specific focus on temperature, though narrow, allowed for ensured diversity to be determined among the functional groups for this specific trait

- Predicted losses in metabolism.
- Future applications using a time series.
- Standardizing thermal rates among different groups, analysis of primary production, and overall, further work on temperature and other parameters is required.

## Article #14 Notes: Fatty Acid Profiles and Production in Marine Phytoplankton

<b>Source Title</b>	Fatty Acid Profiles and Production in Marine Phytoplankton
<b>Source citation (APA Format)</b>	Jónasdóttir, S. (2019). Fatty acid profiles and production in marine phytoplankton. <i>Marine Drugs</i> , 17(3), 151. <a href="https://doi.org/10.3390/md17030151">https://doi.org/10.3390/md17030151</a>
<b>Original URL</b>	<a href="https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6471065/">https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6471065/</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	marine phytoplankton, fatty acids, fatty acid synthesis, PUFA synthesis, EPA, DHA, SDA, environmental effects
<b>#Tags</b>	<a href="#">Basic Outline of Fatty Acid Metabolism</a>
<b>Summary of key points + notes (include methodology)</b>	Fatty acids, particularly unstatured omega 3's and other lipids play an important role in facilitating growth and reproduction in living things. Phytoplankton are some of the few organisms capable of making these molecules themselves, for those molecules to then be transferred by zooplankton as vectors to the rest of the ecosystem. However, variation in fatty acid composition is an area of interest, as this could have ramifications on upper-trophic diversity and nutritional quality. Through conducting a meta-analysis, it is found that the fatty acid composition among seven major phyla does indeed vary. The percent of biomass the fatty acid makes up is also shown to vary. However, due to few past studies having studied the biomass content of fatty acids, the ramifications of the transfer of different compositions of fatty acids cannot be easily projected. The article also notes many parameters that impact fatty acid metabolism rates, noting that more work needs to be done in analyzing the impact of micronutrients. While the general trend is that there shall be an increase in fatty acid production with a corresponding decrease in nutritional quality, the article emphasizes the need to investigate the ramifications of that.
<b>Research Question/Problem/Need</b>	How does the fatty acid concentration vary among different phytoplankton phyla, and what implications does that have on the nutritional quality provided to the upper parts of the trophic pyramid, as well as on the ecosystem as a whole?

Important Figures

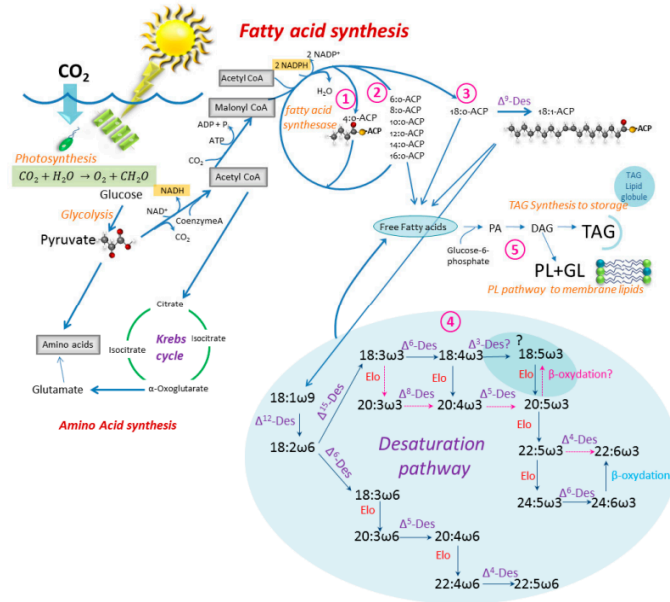


Figure providing background on production of fatty acids in phytoplankton. First, from photosynthesis, glucose is attained, and pyruvate is formed from glycolysis. This pyruvate becomes Acetyl-CoA and travels down various pathways, including Krebs Cycle, and another cycle, where A-CoA gets turned into Malonyl CoA. These two molecules provide the initial carbon for forming fatty acid chain. Subsequently, a series of complex enzymatic reactions occur in the ER whereby chains are elongated and doubly-bonded (i.e., desaturated). Various acids are formed from these processes, and when synthesized, the major fatty acid products are formed. This occurs in the smooth ER.

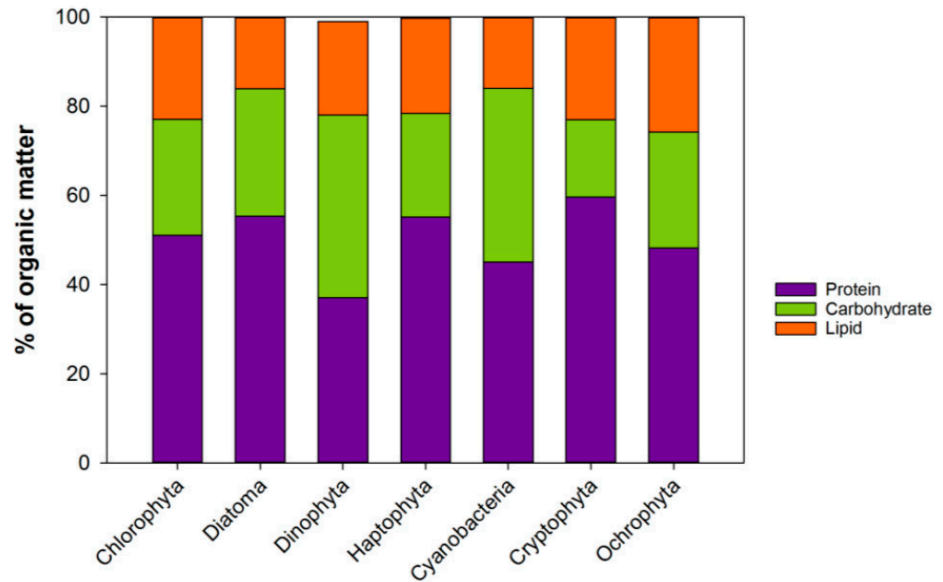
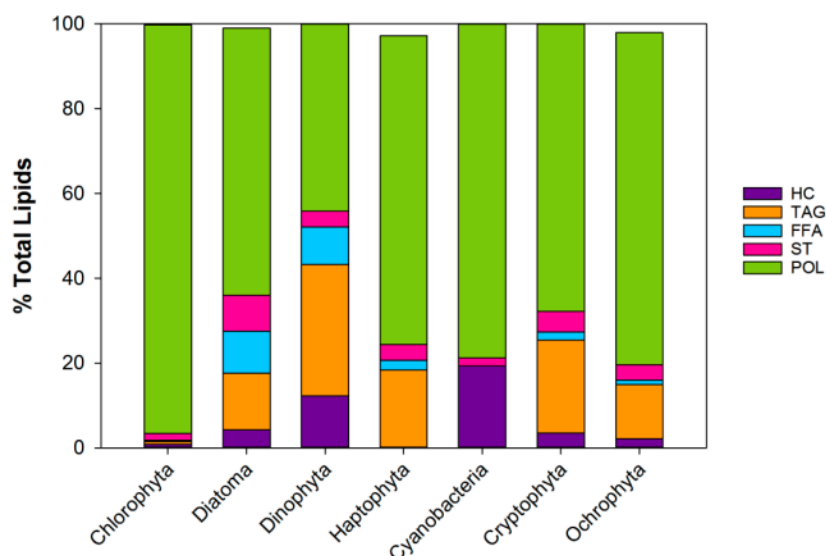


Illustration of proportion of mass of each macromolecule with respect to each phyla of phytoplankton. Proteins make up the plurality of organic matter in all phyla. About 40% -60% of the cells contain protein, with another 20-30%

distributed each to carbohydrates and lipids. Remarkably, there appears to be a consistent ratio of 5:3:2 for protein to carbs to lipids.



This figure illustrates the composition of fatty acids among the seven phyla studied in the meta-analysis, including (from top to bottom in key) hydrocarbons, triacylglycerol, free fatty acids, sterol, and polar lipids. While polar lipids clearly make up the majority (or pluarly fro Dinophtya) of fatty acids in all phyla, the extent to which they do varies from about 45% to 95%. FFAs are notably larger in Diatoma, Dinophyta have the larges TAG concentration, while cyanobacteria the most hydrocarbons. There are many nuances indicating heterogeneity in fatty acid composition.

#### VOCAB: (w/definition)

**Polyunsaturated fatty acids (PUFA)** - Fatty acid molecules (made up of, at the basic level, of hydrocarbon chains) that contain two or more double bonds.

**Highly unsaturated fatty acids (HUFA)** - Fatty acid molecules that contain three or more double bonds.

**Essential Amino Acids (EAA)** - Amino acids that the organism in question is unable to synthesize on their own.

**Galactolipids** - A type of lipid that contains the typical hydrocarbon-chain, that is, the base molecular structure of the hydrocarbon, and in addition, is bonded to a sugar, specifically galactose. Important for metabolic processes.

**Carboxylase** - An enzyme that assists in carboxylation, whereby a substrate is exposed to CO<sub>2</sub>, forming carboxylic acid. It plays a role in more niche aspects of metabolic processes, interacting with amides, alcohols, and other structures.

#### Cited references to follow up on

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	<p>Peters, J.; Dutz, J.; Hagen, W. Role of essential fatty acids on the reproductive success of the copepod <i>Temora longicornis</i> in the North Sea. <i>Mar. Ecol. Prog. Ser.</i> 2007, 341, 153–163.</p> <p>Greenberg, P. <i>The Omega Principle: Seafood and the Quest for a Long Life and a Healthier Planet</i>; Penguin Books: London, UK, 2018</p> <p>Thompson, P.A.; Guo, M.; Harrison, P.J. Effects of variation in temperature. I. On the biochemical composition of eight species of marine phytoplankton. <i>J. Phycol.</i> 1992, 28, 481–488.</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How do the speed and other hydrodynamical characteristics of a water body impact fatty acid production?</li> <li>2. How can the transfer of specific fatty acids be modeled in a neural network of a food chain?</li> <li>3. Given an increase in the concentration of iron and other trace metals, is there a decrease in fatty acid metabolism?</li> <li>4. How do the impacts of carbohydrate metabolism on fatty acid metabolism compare to that of amino acid metabolism?</li> </ol>

## NOTES

### Abstract:

- Phytoplankton sequester carbon, use it to produce macromolecules
- In particular, main source of omega 3 polyunsaturated fatty acids (PUFA), critical nutrient in marine food webs.
- Meta-analysis performed on 160 fatty acid profiles across 7 phyla indicate significant inter-phyla and inter-class differences in the types of fatty acids produced.
- Goes on to list certain acids as being most abundantly produced by given class/phyla
- There are also highly unsaturated fatty acids (HUFA), of which some phyla are better at producing than others
- Important nutritional and environmental impacts on the transfer of these molecules
- Environmental stressors projected to decrease fatty acid movement

### Introduction:

- Carbon integrated from common metabolic pathways, combined w/ phosphorus and nitrogen to create phytoplankton cell
  - Provides the macromolecular nutrients for upper trophic pyramid (crucial in growth, reproduction, health, stability, etc.)
- Zooplankton transfer these molecules → important vectors
- Macromolecular analysis of the different phytoplankton genera to see quality of nutrition of zooplankton, given their role as vectors (holistic ecosystem analysis)
- Some essential nutrients aren't/can't be created by the upper-energy level organisms, which is where phytoplankton become important
  - PUFA omega3s specifically, hence focus on fatty acids.

*Biochemistry Behind these dynamics*



- C is the main element of all compounds and cell structure, binded to different types of molecules in different macromolecular nutrients (quality varies)
- C = indicator of biomass
- N is main building block of the proteins which C is connected to- C/N ratio indicator of nutritional quality + protein structures info.
- See figure 2 analysis above on protein/macromolecular content
- Essential Amino Acid concentrations used to measure protein quality.
- Protein quality found to be homogenous, with significant differences only emerging when considering groups once they have received other nutrient, namely fatty acids.
- W/ regards to carbs, although providing energy, there is little nutritional value for the zooplankton vectors (no trace/nitrogen in molecules)
- With const 5:3:2 ratio among macromolecules, why does nutritional quality success in predators vary?
  - As such, lipids are the focus here
- Background on Glucose → Lipid Metabolism.
  - Crucial for understanding fatty acid comp in phytoplankton
  - Aerobic production methods for fatty acids in ER and chloroplast
  - Pyruvate from glycolysis → pyr becomes Acetyl-CoA (Coenzyme A) → Krebs Cycle, bonding with carboxylase → Malonyl A
  - A-CoA & M-CoA provide C for the fatty acid chain
  - A series of complex enzymatic reactions occur, elongating and desaturating (double bonding) the C chains in the ER.
  - Finally, various acids from the previous steps are synthesized to form the larger-scale macromolecules (e.g. triglycerides, phospholipids, etc.) in smooth ER
- Key here is that phytoplankton have the enzymes that higher-trophic organisms lack to directly produce these essential fatty acids (namely omega3s and 6s as stated by article), hence, phytoplankton are a crucial food source for their predators so they can get these essential but not self-produced molecules.
  - Note: omega is the location of the first double-bond

#### Methodology:

- Meta-analysis performed
- Freshwater species omitted, and the species observed were sorted by Linneaus classification
- Data collated and sorted into categories of “protein”, “carbs”, and “lipids”
- Fatty acid data presented is a percentage of total fatty acids. Homogeneity among the genera and species was analyzed, and the class means taken for each phyla.
- There are discrepancies in the fatty acid data, so to accomodate, any acids with concentrations below 0.1% were not counted or analyzed in the meta-analysis

#### Results:

- Lipid composition varies extremely among phytoplankton (figure 4 analysis above)
- Study attempts to determine the amount of fatty acids found per carbon as a means of understanding how much food value of fatty acids is contained when predation occurs,
- However, there is little data on this subject, hence the mean estimates produced for the phyla in the meta-analysis vary greatly. Nonetheless, it is still clear that the fatty acid content and the amount of nutritional value offered varies greatly.

- Chlorophyta, with regards to fatty acid nutrition, has the worst performance
- Higher temperature results in greater production of fatty acids but reduction in nutritional quality.
- Light is also a crucial component of fatty acid synthesis: elongation and desaturation enzymes rely on light
- Nitrogen and phosphate are also necessary components.
- However, more information is needed on the impact of trace metals on fatty acids.
- All of these parameters play a role somewhere along the chain of reactions that occur for fatty acid metabolism.
- These controls on metabolism can lead to changes in their chemical composition, which important to both (a) understand and (b) manipulate empirically.
- Fatty acids can be used as biomarkers given this heterogeneity.
- The main idea is that varying fatty acid concentrations provides different levels of nutrition to predators.

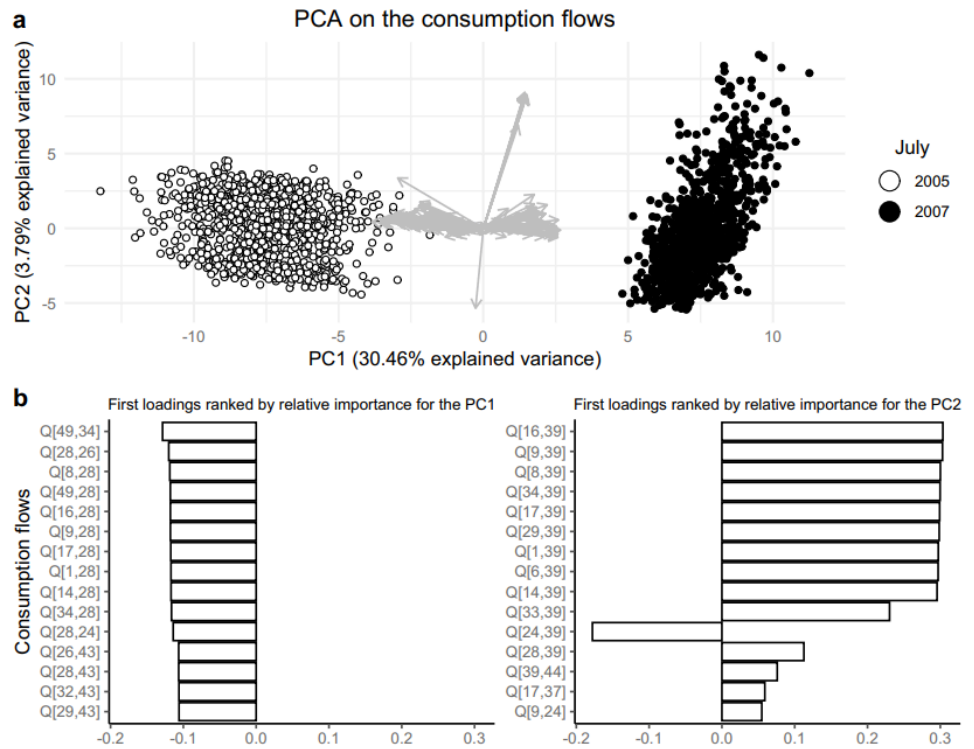
#### Discussion:

- Omega3-lipids are crucial to health, and it is through phytoplankton that it is transferred to the rest of the food chain.
- Major economic demand in fish industry
- Global warming → ocean stratification, warmer temperature, nutrient deficiencies all limit phytoplankton growth.
- Emphasis on the need to understand (model!) the situation as changing dynamics are causing this increasing quantity but decreasing nutritional quality in fatty acids.

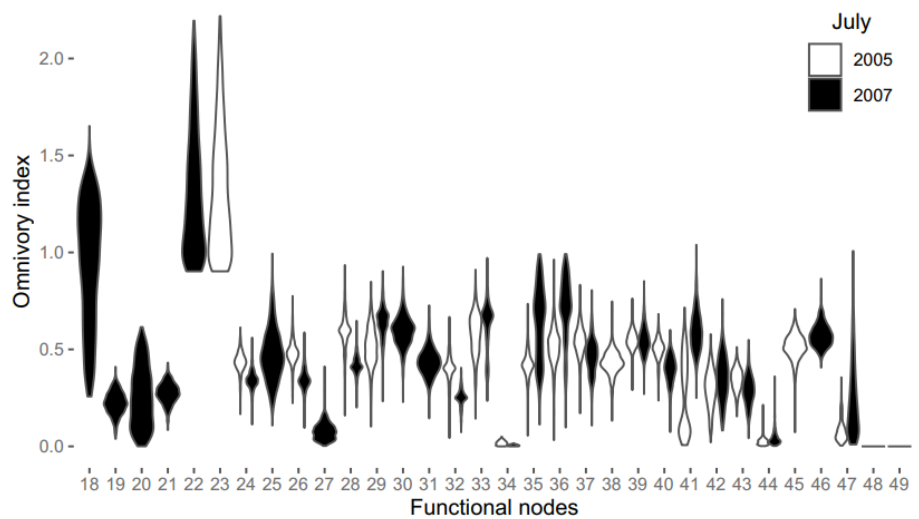
## Article #15 Notes: Planktonic ecological networks support quantification of changes in ecosystem health and functioning

<b>Source Title</b>	Planktonic ecological networks support quantification of changes in ecosystem health and functioning
<b>Source citation (APA Format)</b>	Loschi, M., D'Alelio, D., Camatti, E., Aubry, F. B., Beran, A., & Libralato, S. (2023). Planktonic ecological networks support quantification of changes in ecosystem health and functioning. <i>Scientific Reports</i> , 13(1). <a href="https://doi.org/10.1038/s41598-023-43738-y">https://doi.org/10.1038/s41598-023-43738-y</a>
<b>Original URL</b>	<a href="https://www.nature.com/articles/s41598-023-43738-y">https://www.nature.com/articles/s41598-023-43738-y</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided for this article.
<b>#Tags</b>	<a href="#">Key Attributes of a Food Web</a> <a href="#">Basic Computational Construction of a Food Web</a>
<b>Summary of key points + notes (include methodology)</b>	The goal of this study was to produce a computational food web modeling the transfer of energy for the Venice Lagoon using neural networking. Data from 2005 and 2007 were compared. A neural network was established, with various groups and species assigned nodes, each with their own parameter values. Then, following simulations of the food webs in question, output on ecological conditions were provided. Beyond high resilience and low cycling in 2005, and the opposite situation in 2007, there was not a significant difference found in conditions between the two years. The model was proven to be valid tool usable for future studies.
<b>Research Question/Problem/Need</b>	How do the trophic conditions of different timeframes of the same hyperdynamic water body (specifically Venice Lagoon) differ, and, using ecological network modeling, how can adaptability be characterized, specific roles identified, and inter-parameter interactions better understood?

Important Figures

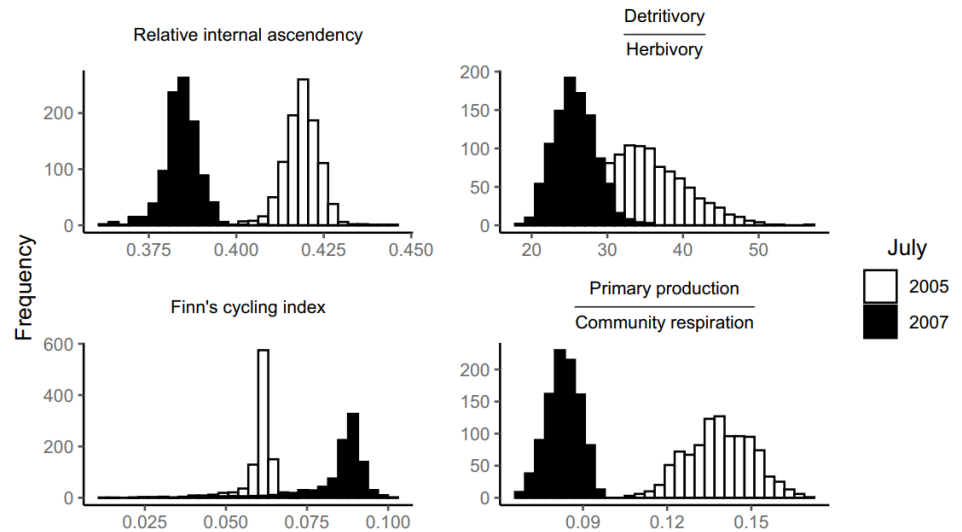


Principal components analysis of the consumption flows, along with the ranks of the most influential nodes is provided by magnitude in the above figure. The white plots represent the resulting datapoints from 2005, and the black ones represent 2007. Each vector represents the impact each node exerts on the model. In conjunction with the supporting bar graphs, it has been found that the nodes containing Strombididae, *Evadne* spp., and *Podon* spp. were the most impactful.



This is a violin plot of the omnivory index, compared between 2005 and 2007. In line with the findings of this study, there was not a significant

change in this ecological attribute between 2005 and 2007. The centers and spreads are mostly the same across both years, as seen visually above. There are some differences, however, they are not enough to be considered significant.



These histograms are of the frequency among the multiple samples collected of the various ecological characteristics measured as outputs of the food web. It performs a similar function as the above figure, only using a histogram as opposed to a violin plot. Here, there seems to be a few more differences between '05 and '07, indicating that there are *some* changes between the two years, even if they are not completely significant. It seems that 2007 saw less resiliency (internal ascendancy) but more cycling, whereas the opposite is true for 2005. From 2005 to 2007, it seems that community respiration increased, as did feeding on producers.

#### VOCAB: (w/definition)

**Turnover Rate** - Speed with which an enzyme is able to operate.

**Monte Carlo Random Sampling** - A method involving repeated random sampling whereby numerical values are attained. The context is often to find some deterministic factor or trend.

**Functional Nodes (FNs)** - A "point" on a neural network

**Violin Plot** - Compares probability distributions of given factor or factors. It acts a broader form of a boxplot. Instead of just plainly having the median and quartiles, it also contains the distribution of the numbers themselves. It's basically just a histogram layed on top of a boxplot.

**Mixotrophic** - An organism whose means of energy attainment lie on a spectrum in between autotrophs and heterotrophs.

**Detritivores** - Organisms that mainly feed on detritus, dead organic matter.

#### Cited references to follow up on

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	<p>Ulanowicz, R. E. &amp; Kay, J. J. A package for the analysis of ecosystem food networks. <i>Environ. Softw.</i> 6, 131–142 (1991)</p> <p>Christensen, V. &amp; Pauly, D. ECOPATH II—a software for balancing steady-state ecosystem models and calculating network characteristics. <i>Ecol. Model.</i> 61, 169–185 (1992).</p> <p>D’Alelio, D., Hay Mele, B., Libralato, S., Ribera d’Alcalà, M. &amp; Jordán, F. Rewiring and indirect effects underpin modularity reshuffling in a marine food web under environmental shifts. <i>Ecol. Evol.</i> 9, 11631–11646 (2019).</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Can the computational techniques employed for this model be applied to other ecosystems?</li> <li>2. How does the number of nodes, i.e. ecosystem complexity relate with the fit of a neural network food web?</li> <li>3. How can the impact of environmental parameters that impact food web dynamics be incorporated into this computational model?</li> <li>4. What tools can be used to measure phytoplankton dynamics in real time?</li> </ol>

## NOTES

### Abstract:

- Changes in aquatic changes impact metabolic rates and energy transfer of phytoplankton
- Good indicator of ecosystem health
- Water body in Venice Lagoon had food web data between July 2005 and July 2007 compared.
- Monte Carlo random sampling of parameters used to derive 1000 food webs
- Certain species of phytoplankton were found to be important in the splitting and controlling food web (controlling energy transfer and all that)
- Slight variations in functional nodes (FNs), but overall same results between the 2 years
- Consumer use leads to greater sediment resuspension, increasing turbidity,
- Main source of primary production = benthic FNs/organisms
- Food web maintained equilibrium, although environmental stress caused efficiency to be sacrificed in favor of resiliency.

### Introduction:

- Dif. parameter values impact productivity and tax comp
- Particularly, the turnover rate, i.e., rate at which enzyme operates is impacted, making phytoplankton communities good indicators of ecosystem health
- Dif. phytoplankton dynamics = dif. Trophic structure
- Various processes, including mixotrophy, heterotrophy, and detritivory are used to facilitate rapid adaptation to environmental change
- Helps maintain resilience and basic processes of equilibrium state
- Transitional coastal lagoon is a good case study: area has a lot of variation.

- E.g. freshwater/seawater, hydrodynamics, anthropogenic influences, etc.
- These factors impact energy transfer among food web, changing ecological comp
- In observing how phytoplankton are changing can indicate ecosystem resilience, and importantly, the mechanisms behind that.
- Herein lies the utility of ecological network models. Due to their holistic nature that captures the line-by-line complexities, analysis can reveal impact of changes on ecosystem operation.
- The reason to focus on phytoplankton is due to their role in critical microbial and energy transfer mechanisms.
- Coastal area study is particularly useful: more data availability + looking at a situation where multiple parameters change.
- Venice July '05 vs. July '07 comparison of trophic conditions performed. July was chosen b/c it is when the biomass peaks, and as a corollary, when more data on wider variety of zooplankton is present, the occurrence of important ecological processes.
- Goal in this analysis is to identify specific roles among groups of phytoplankton, as well as compare trophic state to assess factors and their impact and implications at two dif. Timeframes.
- Innovative iterative random samplings of parameters approach was taken to construct the food webs.

#### Methodology:

- Venice Lagoon is part of the Italian Long-Term Ecological Research Network. Large water body, transitional, well-studied. Majorly important in socio-ecology,
- Northern area, Palude della Rosa, was studied, notable for salt (seawater, and this is the highly saline Mediterranean) and freshwater (river discharge) inflow.
- All sampling collected during specific tide times to avoid biasing data; July 2005 and Jul 2007 used (tax com., biomass in mg C m<sup>-3</sup>).
- Various types of phytoplankton and other related species (bacteria, zooplankton) were sampled/had specific laboratory procedure for testing
- FNs generated for each species at lowest tax classif possible: grouping together to form nodes and such based off similar ecological function, each FN contains size and role.
- Inter-species pref vary ⇒ only two params (benthic/pelagic preference)
- Parameters:
  - $\mu$  → production rate per biomass unit (d<sup>-1</sup>)
  - $\alpha$  → consumption rate per biomass unit (d<sup>-1</sup>)
  - $\epsilon$  → unassimilated fraction of biomass consumed
  - $ph$  → phototrophy proportion in individual metabolism (0 = heterotroph, 1 = autotrophs, and in between for mixotrophs)
  - $\gamma$  → Proportion of flows to non-living nodes (e.g. feces death etc)
  - Properties range given the organism of the FN
  - Ordinal variables describing strength of interaction 0 (none) to 3 (strong)
- Assumption: Balance between production, mortality and predation:

$$\mu_k \cdot B_k - \sum_{j=1}^n (\alpha_j \cdot B_j \cdot DC_{k,j}) - m_k = 0$$

- $\mu_k$  = production rate per biomass unit;  $B_k$  = biomass;  $m_k$  = mortality;  $j$  is the index of the properties of the predator (consumption, biomass, and  $DC_{k,j}$  is proportion of the prey in diet of the predator)
- Balanced model used for the non-living nodes:

$$\sum_{i=1}^n [\gamma_{i,d} \cdot (\epsilon_i \cdot \alpha_i \cdot B_i + m_i)] - \sum_{j=1}^n (\alpha_j \cdot B_j \cdot DC_{d,j}) - ex_d + im_d = 0$$

- $\epsilon_i$  = unassimilated fraction of biomass consumed. The first summation is flow of energy of living node to nonliving one. Subtract from that the consumptions of the detritivores, as well as the export ( $ex_d$ ). Add the  $im_d$ , the import.
- Estimation of respiration

$$R_i = Q_i - P_i \cdot (1 - ph_i) - UN_i$$

- $Q$  = consumption,  $P$  = production,  $ph$  = phototrophy proportion for node  $i$ ,  $UN$  = unassimilated material
- The above parameters for each FN was generated from taking random sample for the possible range.
  - Use of the ordinal variable strength 0-3 here; between 0 and 1 (minmax normalization), two bounds for each parameter 0 to 1 were set.
    - 1: 0 to first; 2: 1st to 2nd; 3: 2nd to 1
- All FNs had a system of linear equation containing the range of the parameters. Other parameters not incorporated in model calculated by those that were.
- No specific values/solutions to the equation; rather, random values for the parameters were used by taking repeated random sample from the possible range
- Another assumptions made:

$$R_i \geq 0$$

- $0.15 \leq GE_j \leq 0.5$ 
  - Where  $R_i$  is the respiration flow and  $GE$  is the gross food conversion efficiency, ratio between heterotrophic production/consumption
- Various ecological attributes of the food web calculated for both '05 and '07 and statistical comparison was performed (formulae can be found in the article)
  - Relative Ascendency: The ability of an ecosystem to be resilient given disturbances due to sheer size and organization. Measure between efficiency and resilience (0 = min efficiency/max resilience, 1 the opp)
  - Relative internal ascendency: The same measure described above, based on the internal transfer of energy (same scale)
  - Finn's cycling index: The proportion of flows attributable to cycling (0 = no cycling, 1 = all material cycled)
  - Ratio of detritivory to herbivory: Comparison between consumption of producers vs. nonliving nodes;  $>1 \rightarrow$  more nonliving nodes;  $<1 \rightarrow$  more producers;  $1 \rightarrow$  about equal



- Ratio of primary production to community respiration: During ecological succession,  $\neq 1$ ; during equilibrium,  $=1$
- Omnivory index: Variation of trophic levels of a consumer's prey (closer to 0 the more specialized in feeding)
- PCA used on only the non-constant flows of different sizes from '05 to '07
  - Two largest vectors == nodes most significant to the split of the food web
- Mood's Median Test and other nonparametric approaches taken (parametric conditions not met)
- R used as a programming language

Results:

- See figure analysis above

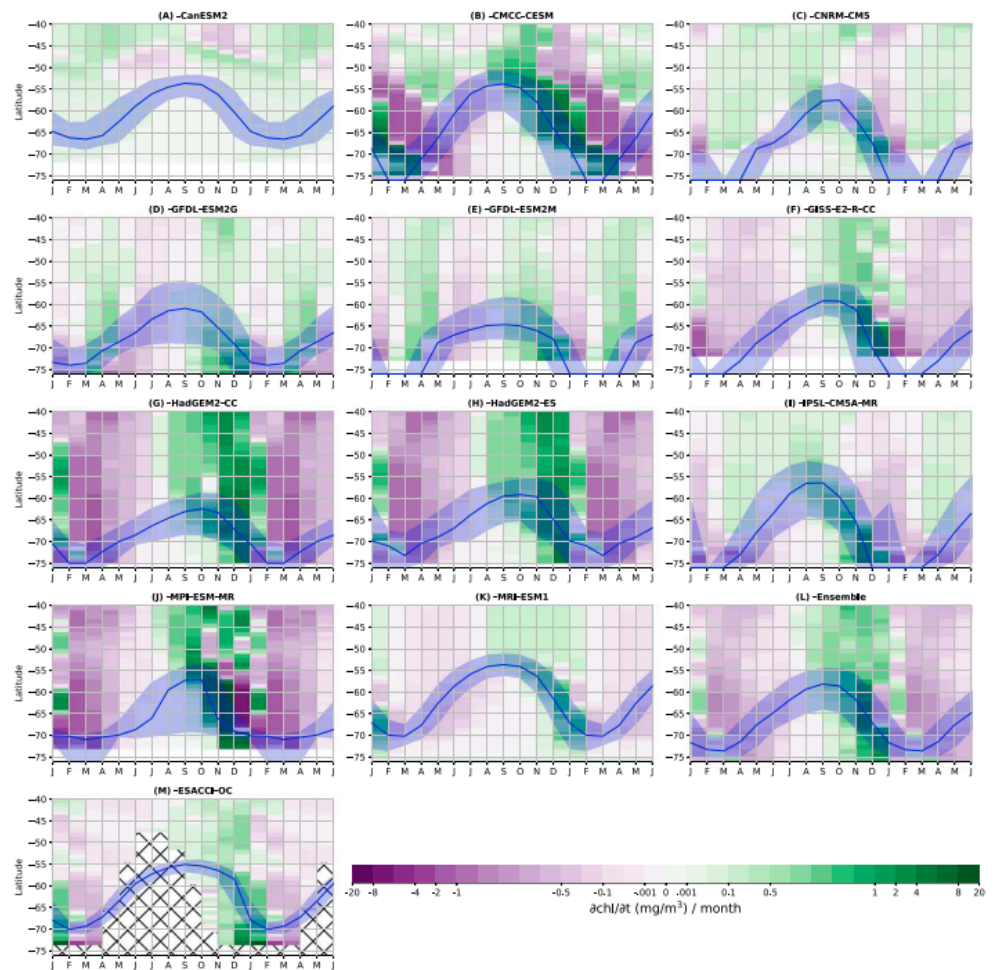
Conclusions

- Lack of significant ecological difference between '05 and '07
- Valid computational food web with intriguing and accurate results successfully produced.

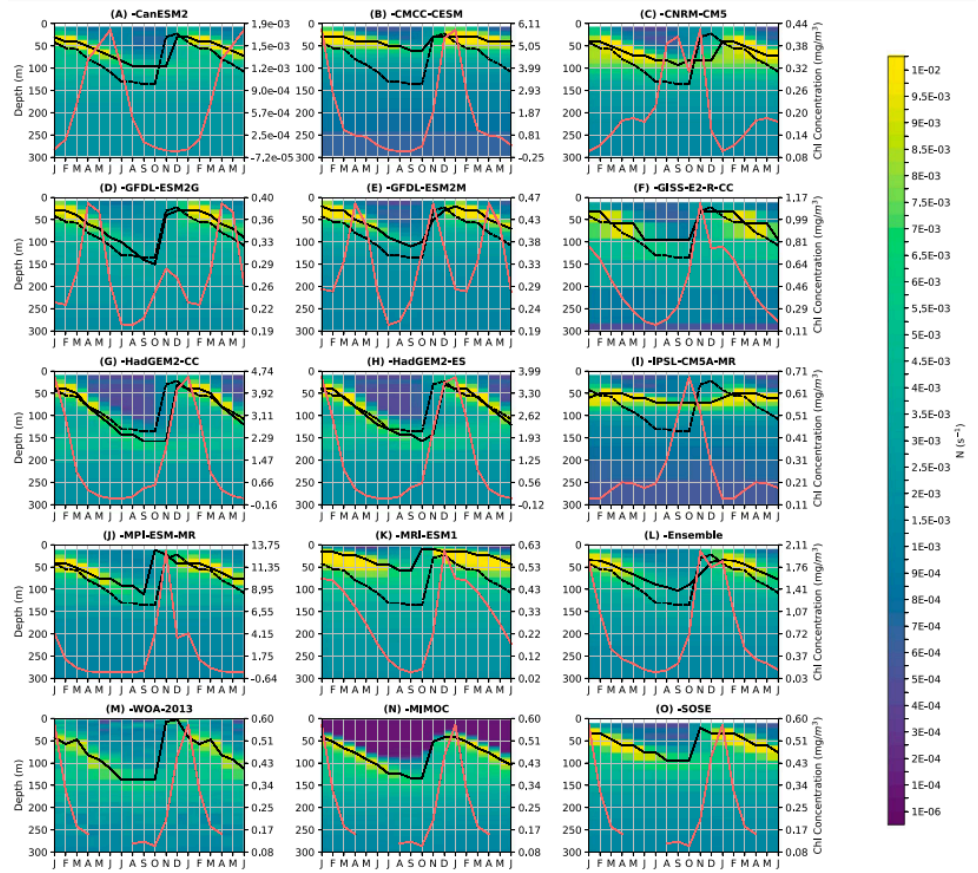
## Article #16 Notes: A Link Between CMIP5 Phytoplankton Phenology and Sea Ice in the Atlantic Southern Ocean

<b>Source Title</b>	A Link Between CMIP5 Phytoplankton Phenology and Sea Ice in the Atlantic Southern Ocean
<b>Source citation (APA Format)</b>	Hague, M., & Vichi, M. (2018). A Link Between CMIP5 Phytoplankton Phenology and Sea Ice in the Atlantic Southern Ocean. <i>Geophysical Research Letters</i> , 45(13), 6566–6575. <a href="https://doi.org/10.1029/2018gl078061">https://doi.org/10.1029/2018gl078061</a>
<b>Original URL</b>	<a href="https://agupubs.onlinelibrary.wiley.com/doi/full/10.1029/2018GL078061">https://agupubs.onlinelibrary.wiley.com/doi/full/10.1029/2018GL078061</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided for this article.
<b>#Tags</b>	<a href="#">Relevant Instances of Computational Limitations</a>
<b>Summary of key points + notes (include methodology)</b>	The aim of this article is to evaluate a energy inaccuracy that the CMIP5 has in simulating Southern Ocean dynamics. The model simulates the consequences of CMIP5 failing to account for the fact that deep winter mixing prevents biomass accumulation until late spring via light limitation. It is seen that there are biases in proper chlorophyll a concentrations, poleward bias of the Marginal Ice Zone line prediction, shallower mixing layer, and earlier bloom periods. It is concluded that more physical constraints and other modifications need to be applied to the CMIP5 model to increase model validity.
<b>Research Question/Problem/Need</b>	What impact does the inaccurate modeling of the mixing layer depth in the Atlantic southern ocean by the CMIP5 model have on its simulation of phytoplankton dynamics in this region?

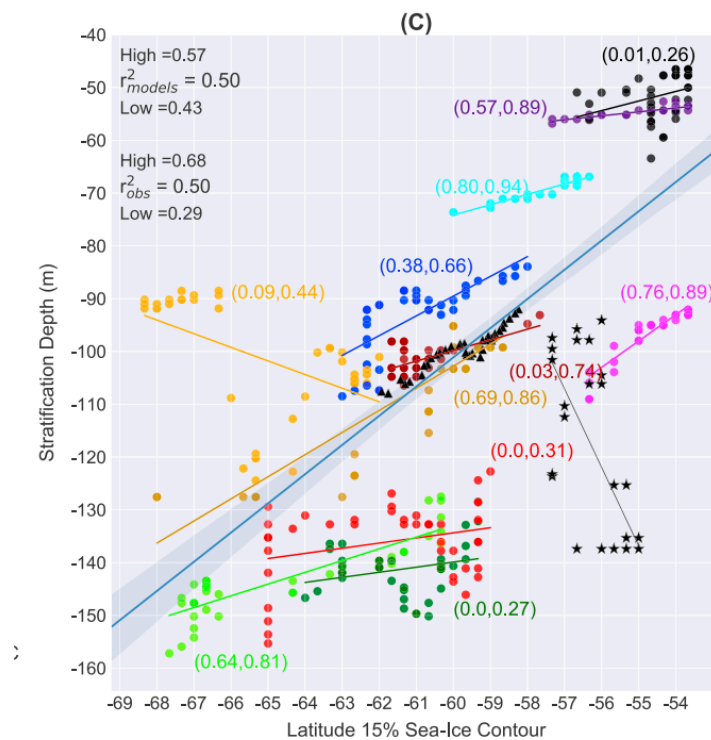
## Important Figures



This figure contains predictions from different models on the location of the boundary of the marginal ice zone, with shading representing the error. Greener colors indicate areas of higher chlorophyll concentration, whereas purpler ones indicate areas of lower concentration. Latitude is used as the y-axis. The nature and location of the peaks and troughs differs across all models. There is a bias towards shallower depths.



Here, circulation in the ocean currents is measured by the various models. The red and black lines on top represent the chlorophyll concentration and stratification depth respectively. Again, the predictions of these measurements varies across all of the models.



The figure depicts the relationship between stratification depth and ice edge location among the models. Overlain in blue is the linear regression with the 95% confidence intervals (bootstrapped used) shaded in light blue. R<sup>2</sup> values are provided, as well as the high and low confidence intervals shown by the shading. Each data point shown represents a different degree of longitude for each model. Notice the lack of cluster in the dots among each model; this indicates that there is improper predictions about the location of the stratification layer.

**VOCAB: (w/definition)**

**Isopycnal** - Ocean layers that stratified on the basis of density  
**Hovmöller Plot** - A unique way of modelling oscillatory meteorological data, typically on a temporal scale.  
**Contour** - A series of curves (based on function of two variables) drawn to represent a 3D space such that such that function values remain constant. Lines are drawn along the areas of common z-scores.  
**Brunt-Väisälä Frequency** - The stability fluids are able to maintain when exposed to vertical perturbation in currents.

**Cited references to follow up on**

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Anav, A., Friedlingstein, P., Kidston, M., Bopp, L., Ciais, P., Cox, P., et al. (2013). Evaluating the land and ocean components of the global carbon

	<p>cycle in the CMIP5 earth system models. <i>Journal of Climate</i>, 26, 6801–6843. <a href="https://doi.org/10.1175/JCLI-D-12-00417.1">https://doi.org/10.1175/JCLI-D-12-00417.1</a></p> <p>Boyd, P. W. (2002). Review of environmental factors controlling phytoplankton processes in the Southern Ocean 1. <i>Journal of Phycology</i>, 38(October 2001), 844–861. <a href="https://doi.org/10.1046/j.1529-8817.2002.T01-1-01203.x">https://doi.org/10.1046/j.1529-8817.2002.T01-1-01203.x</a></p> <p>Dunne, J. P., John, J. G., Shevliakova, S., Stouffer, R. J., Krasting, J. P., Malyshev, S. L., et al. (2013). GFDL's ESM2 global coupled climate-carbon Earth system models. Part II: Carbon system formulation and baseline simulation characteristics. <i>Journal of Climate</i>, 26(7), 2247–2267. <a href="https://doi.org/10.1175/JCLI-D-12-00150.1">https://doi.org/10.1175/JCLI-D-12-00150.1</a></p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Are there any other locations in the world that are improperly modeled, and if so, how does that impact phytoplankton simulations?</li> <li>2. How does the use of a time time series impact the accuracy of a climate model?</li> <li>3. How can historical data be better utilized in models of future projections so as to reduce inaccuracy?</li> <li>4. How do varying level in chlorophyll a concentration, or any other parameter</li> </ol>

## NOTES

### Abstract:

- Bias/data problem in Coupled Model Intercomparison Project Phase 5 (CMIP5).
- Inaccurate depiction of late winter primary production
- Benthic/Sub-pelagic light availability is correlated with early growth, as winter mixing and ice cover prevent growth, dynamics which go unmodeled.
- Correlation between stratification and the extent of sea ice. Increased stratification (particularly in late winter) = ice extent ^ (b/c closer to the equator)
- Given the correlation stated above, and how well models control for it (including each internal factor), models were sorted into groups.
- Models with less bias had better predictions (even when having proper control of one factor only)

### Introduction:

- CMIP5 models on Antarctic sea ice trends have been known to be inaccurate, the implications of which have yet to be fully investigated
- Specific focus is on impact of the inaccuracy on phenological phytoplankton modeling in the Atlantic Southern Ocean (Atlantic b/c largest Chlorophyll concentrations and large spatial variability)
- Focus on the marginal ice zone, where ice exists in some degree but atmosphere-oceanic exchange is still possible. This area is known for greater primary production

- As ice melts, the water column stabilizes, with a pelagic layer of freshwater forming that allows for the biomass of phytoplankton to increase. But there is also iron and other factors that CMIP5 doesn't account for.
- Many past studies have indicated that the CMIP5 predicts the location of the mixing layer to be shallower than what is actually the case, indicating that there was too much freshwater being input into the model
- Leads to a lack of mixing → impacts the light/nutrient conditions, changing results (so let's see how those change, based off the inaccurate vs. accurate estimate of mixing layer)

#### Methodology:

- 11 CMIP5 models on historical simulations were collected (1976-2005 data timeframe)
- Variables considered: pelagic chlorophyll mass concentration, sea ice area fraction, salinity, and potential temperature
- Various satellite models were tested and compared
- Data spreads from 20°W to 20°E
- Marginal Ice Zone (MIZ) defined as area of 15-80% ice concentration contours; 15% is the northern edge; to account for different definitions among models, a common region of analysis was used in all models: 58°S to 62°S
- Allows for meaningful comparisons to be performed
- Main measure here is the temporal prediction of bloom initiation and peak, defined when the chlorophyll concentration passes 5% greater than the median; bloom peak = chlorophyll peak

#### Results:

- See figure analysis above
- Poor modeling of chl a concentrations has been supported by evidence
- There is a bias towards shallower depths.

#### Conclusion:

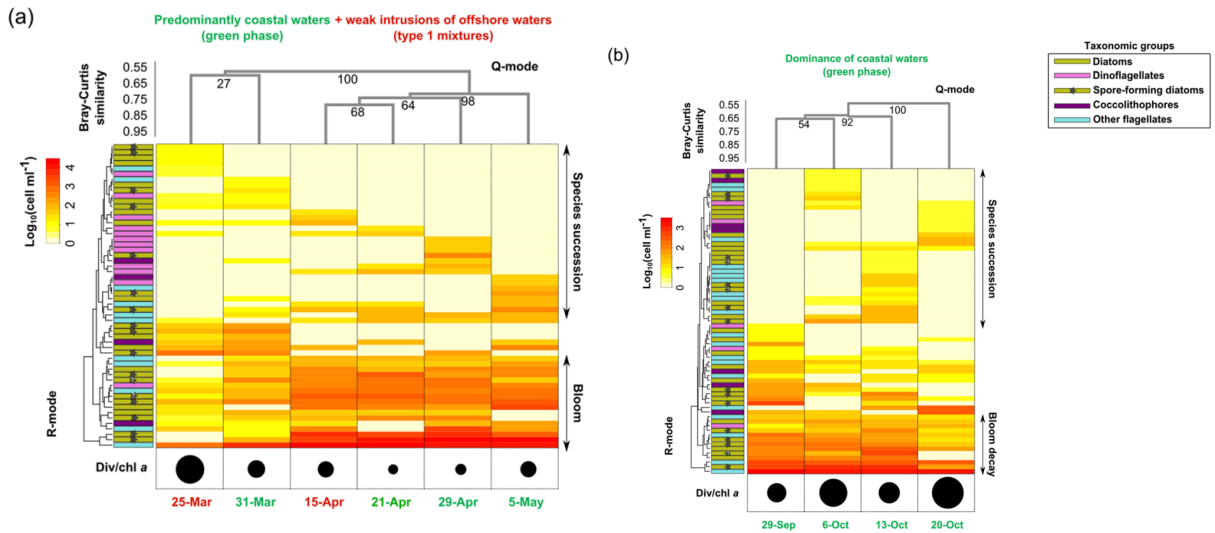
- All models are assorted into groups and analysis of them is performed. The findings show clear biases in chl a concentration, poleward bias of the MIZ line, shallower mixing layer, and earlier bloom periods.
- In the real ocean, ice cover, deep winter mixing, and associated light limitation prevent biomass accumulation until the late spring. This is not present in the CMIP5

## Article #17 Notes: Disentangling physical and biological drivers of phytoplankton dynamics in a coastal system

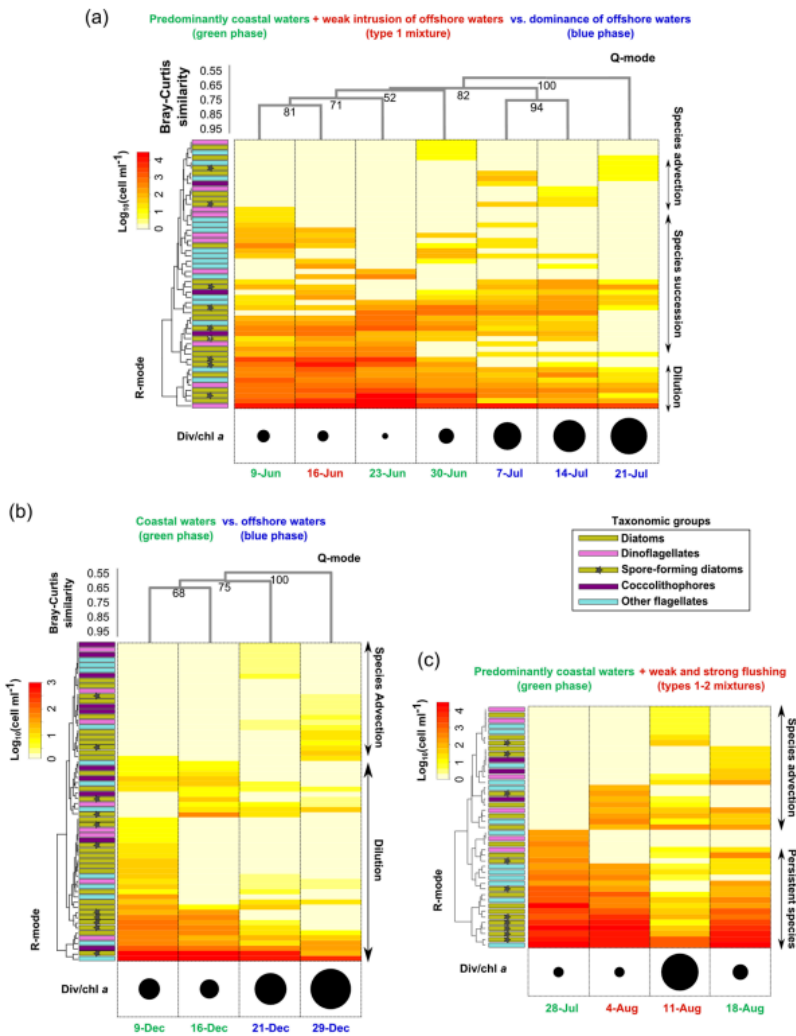
<b>Source Title</b>	Disentangling physical and biological drivers of phytoplankton dynamics in a coastal system
<b>Source citation (APA Format)</b>	Cianelli, D., D'Alelio, D., Uttieri, M., Sarno, D., Zingone, A., Zambianchi, E., & d'Alcalá, M. R. (2017). Disentangling physical and biological drivers of phytoplankton dynamics in a coastal system. <i>Scientific Reports</i> , 7(1). <a href="https://doi.org/10.1038/s41598-017-15880-x">https://doi.org/10.1038/s41598-017-15880-x</a>
<b>Original URL</b>	<a href="https://www.nature.com/articles/s41598-017-15880-x">https://www.nature.com/articles/s41598-017-15880-x</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided for this article.
<b>#Tags</b>	<a href="#">Potential Data Analysis Tool (Analogus to December Fair)</a>
<b>Summary of key points + notes (include methodology)</b>	This study aims to examine the interaction between physical and ecological parameters as they impact phytoplankton dynamics in the Gulf of Naples. A novel high frequency coast radar system was used to track water movement as well as the movement of various patches of phytoplankton, using Lagrangian Modelling methods. The results from this computer simulation were connected with ecological parameters of chlorophyll a and salinity. Complex relationships between these two areas, as well as with phenology were found. Overall, summer is the most abundant season, whereas winter is the least; meanwhile, throughout the year, diatoms are the most abundant, dinoflagellates the least. This study successfully differentiated between the biological and hydrodynamical forces at play, developing a methodology applicable to other areas.
<b>Research Question/Problem/ Need</b>	How can novel high frequency coastal radars (HFRs) be used to effectively model and differentiate physical and biological parameters impacting phytoplankton, and how can a methodological framework for these ends be developed?



Important Figures



These figures illustrates heat maps of specific patches of phytoplankton that were tracked by the HFRs. The specific dates indicate when the data was collected. The warmer colors indicate greater biomass (in terms of log of cell abundance). Analyzing the corresponding species and the biomass, it seems that non-sporal diatoms are the most abundant, whereas the the dinoflagellates are the least abundant. This is for spring and autumn,



Overall, in the winter (looking on the left on the bottom), the diatoms are more abundant, but overall, their abundance is lower, whereas the opposite is true in summer: abundance increases even more, even among the less abundant groups.

**VOCAB:**  
(w/definition)

**Lagrangian Modelling** - A series of methods and models where points of a moving surface with inconsistent flux and other dynamics can be modeled.

**Heat Map** - A method of data display whereby two dimensions and their levels are placed into a 2D array. Each cell has a sort of hue along a color gradient to represent its corresponding value across the spectrum of parameter that is being measured.

**High frequency coastal radar** - A tool that is able to measure the velocity of pelagic ocean currents live.

**Allogenic** - Impacts due to nonliving factors.

**Cited references to follow up on**

D'Alelio, D., Libralato, S., Wyatt, T. & Ribera d'Alcalà, M. Ecological-network models link diversity, structure and function in the plankton food-web. Scientific Reports 6, 21806, <https://doi.org/10.1038/srep21806> (2016).

Hammer, Ø., Harper, D. A. T. & Ryan, P. D. PAST: paleontological statistics software package for education and data analysis. *Palaeontologia Electronica* 4, 9–18 (2001).

Ribera d'Alcalà, M. et al. Seasonal patterns in plankton communities in a pluriannual time series at a coastal Mediterranean site (Gulf of Naples): an attempt to discern recurrences and trends. *Scientia Marina* 68(Suppl. 1), 65–83, <https://doi.org/10.3989/scimar.2004.68s165> (2004).

Schapira, M., Vincent, D., Gentilhomme, V. & Seuront, L. Temporal patterns of phytoplankton assemblages, size spectra and diversity during the wane of a *Phaeocystis globosa* spring bloom in hydrologically contrasted coastal waters. *Journal of the Marine Biological Association of the United Kingdom* 88, 649–662, <https://doi.org/10.1017/s0025315408001306> (2008).

### Follow up Questions

1. Similar to how there are tangled interactions between physical and biological parameters present that are observed in this study, are there other fields that exert intertwining impacts?
2. What attributes of a computer model make it universally applicable versus specific to the area is modeling?
3. How can a hierarchical linear model or other similar tool be integrated into a model such as this one, one which takes into account multiple parameters of different fields?
4. How do interfield parametric interactions differ when in connected water body system versus a closed water system?

### NOTES:

#### Abstract:

- Surface current and time series data both analyzed to help identify driving parameters of dynamics
- Species succession was controlled by biological factors (inter-species interaction, physiology, etc.)
- Physical factors were also important (e.g. coastal movement, mixing), as offshore species are integrated into the ecosystem, regulating competition and dominant species
- Findings show that coastal communities are resilient
- A proof-of-concept design to apply to other coastal ecosystems (unifying the heterogeneity among coastal areas)

#### Introduction:

- Allogenic (physical) and autogenic (processes) both regulate changes in phytoplankton
- Physical = mixing, movement, etc.; Biological = Ecological interactions, cytological and other internal processes, etc. + shallower areas have more complex gradient → difficult to understand.
- Important to consider for phytoplankton blooms, which can sometimes be toxic (algae).
- Particular problem with oversimplified analysis (as well known for this project)
- Tools exist to model these two major groups of factors: ecological information and surface circulation, modeled by ever-more-robust high frequency coastal radars (HFRs)
- Lagrangian models have yet to be applied well to marine environments
- This study combines this novel observation of the water currents with ecological site data collected.

- A proof-of-concept study focused on establishing a methodology for future investigations to follow.
- The HFRs produced high resolution observation of the biological and physical parameters, allowing easier connection to phytoplankton dynamics.

#### Methodology:

- Complex description of the HFR calibration is provided
- 2009 was used as year to study as it had the most radar and biological data.
- On Lagrangian Modeling:
  - Surface velocity used as input
  - 10 minute interval tracking movement retrospectively (backtracking sims)
  - 15% error used
  - Particles released in the simulation, with the initial location being determined by result of backtracking sim (accurate up to 4 days)
  - Probability model used to define spatial variability of the origin zones
  - Entry time (shift focus to temporal, not spatial scale), center of mass and the variability of that (this is spatial) was measured.
  - The origins points were designated into sectors, 1-4 and 8 being coastal, 5-7 offshore
- Positions had their corresponding salinity, chlorophyll a measured. There was element of phenologic comparison to this.
- Z-standardization used among these different timeframes to compare the measures in this area (so there was time the particle was located at a given position which had these parameter values which are now being compared)
- Chlorophyll a values determined from direct extraction
- Salinity/chlorophyll a cluster analysis to see the ecological characteristic of the lake studied (Gulf of Naples) (R was used)
  - Bray-Curtis similarity, logarithmic abundance data, alpha index of diversity
    - Independent of n
- Gulf of Naples = area of study
- Complex benthic properties → complex hydronamic properties; seasonal shift between moderately eutrophic and oligotrophic
- Data has been collected very regularly on this body of water (weekly since '84)
- Ratio between alpha diversity and biomass was also considered.
- All of these processes were to help disentangle biological and physical parameters

#### Results:

- Phenological variation in the speed of the water currents.
- Patches of phytoplankton concentrated closer to the coast, sporadic as distance increases
- Various complex interactions between the hydrodynamic and ecological parameters were observed.
  - Phenological variation in what is a driving factor, what is not. Location, chlorophyll a and salinity driving factors in various cases, not in others, etc.
- See analysis of trends in the biomass in figures above

#### Conclusion:

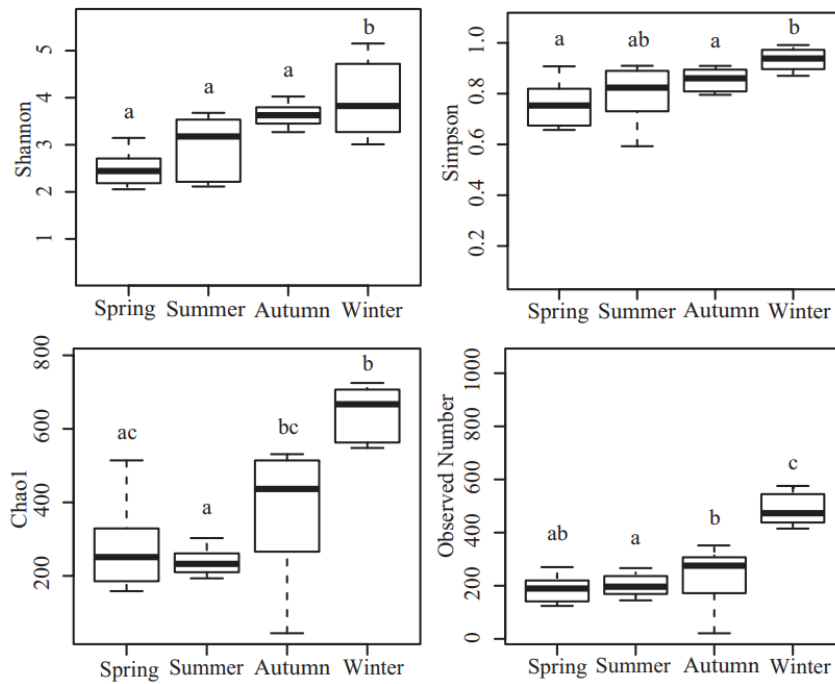
- The computational system was able to successfully allow for the physical vs. ecological parameters in different situations to be determined.

- The farther the origin zone of offshore waters, the larger the modification induced in the phytoplankton community.
- Successful displays of the interaction between biological and physical parameters, and the connection with those dynamics to the dual-impact of the coast and offshore currents.
- Proof-of-concept study applicable to other areas.
- Helpful for management decisions.

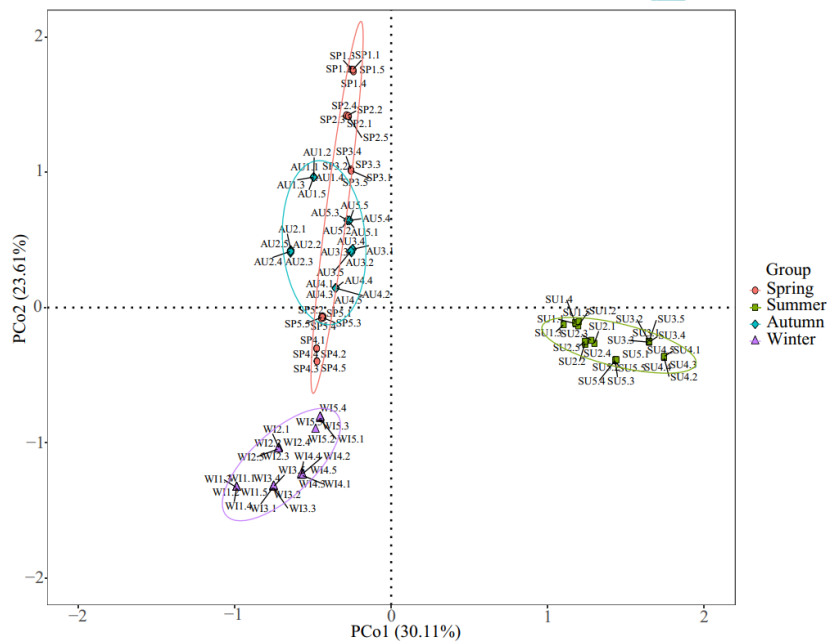
## Article #18 Notes: Ecological stoichiometry influences phytoplankton alpha and beta diversity rather than the community stability in subtropical bay

<b>Source Title</b>	Ecological stoichiometry influences phytoplankton alpha and beta diversity rather than the community stability in subtropical bay
<b>Source citation (APA Format)</b>	Xu, Q., Huang, M., Shu, Y., Li, X., Zhao, H., Tang, J., Jiang, G., Li, Z., Huang, Y., Dong, K., Huang, L., & Li, N. (2022). Ecological stoichiometry influences phytoplankton alpha and beta diversity rather than the community stability in subtropical bay. <i>Ecology and Evolution</i> , 12(9). <a href="https://doi.org/10.1002/ece3.9301">https://doi.org/10.1002/ece3.9301</a>
<b>Original URL</b>	<a href="https://onlinelibrary.wiley.com/doi/full/10.1002/ece3.9301">https://onlinelibrary.wiley.com/doi/full/10.1002/ece3.9301</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Beibu gulf, community stability, distribution, ecological stoichiometry, phytoplankton diversity, rbcL gene
<b>#Tags</b>	<a href="#">Means of Measuring Ecological Diversity</a>
<b>Summary of key points + notes (include methodology)</b>	The purpose of this study is to analyze the parameters that impact phytoplankton diversity. The Beibu gulf was used as a case study, with micronutrient and other relevant data being collected across a large spatio-temporal scale, an entire year of the four seasons, which was novel. The relationships between various indices of diversity and community structure, the temporal patterns of these indices, and their relationship with nutrient stoichiometry were investigated. To this end, PCR and other genomic techniques were used to analyze the diversity. ANOSIM, ANOVA, PoCA, Bray-Curtis dissimilarity, among other statistical tools were used for analysis. It was found that, due to the temperature drop in the spring, this season was of the least abundance and diversity, and every seasons thereafter, successively, had greater value of these metrics. More notably, a significant positive correlation between nutrient stoichiometry and the diversity indices was established. While the findings are convincing, more analysis into the underlying factors observed is needed to develop the bigger picture.
<b>Research Question/Problem/Need</b>	What factors impact phytoplankton community structure and diversity as can be observed from water samples from the Beibu Gulf? Specifically, does ecological stoichiometry play a role, and is there a correlation between community diversity and stability?

**Important Figures**

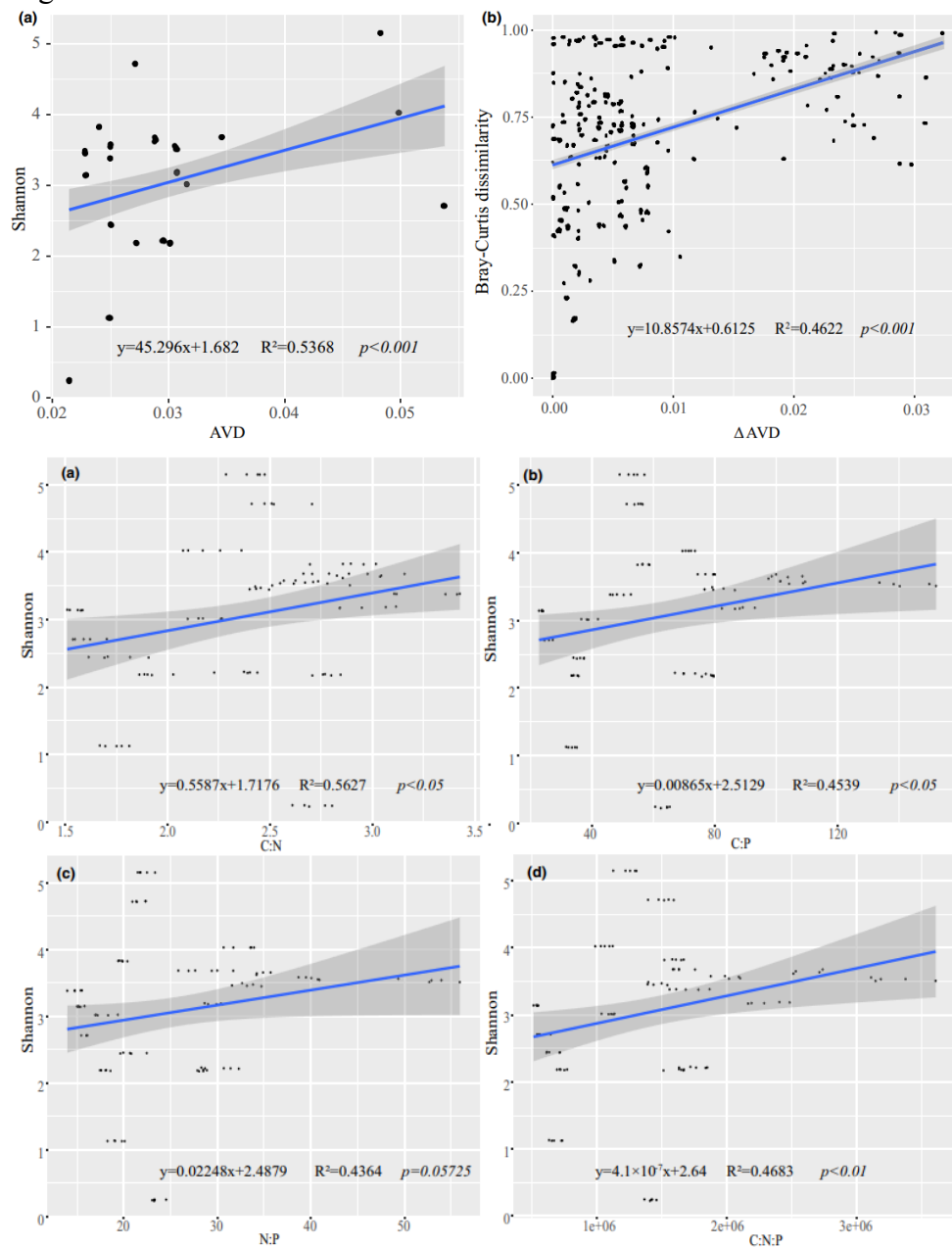


This figure depicts the phenological trends of the alpha diversity indices in the Beibu gulf. These include the shannon, chao1, simpson indices, as well as the raw abundance. Interestingly, it seems that the indices, as well as the abundance, is highest in the winter, followed by the autumn, summer, and spring. Though, some indices have skewed distributions, for example, especially for chao1.



This is a principle coordinate analysis chart of the diversity of the phytoplankton among the four seasons. Given the distances of the groups from the origin, it would seem that there is a

significant divergence for the summer group, as well as the winter group. However, the significance of the variation might be lower for all and autumn, as they are closer to the origin.



From left to right and top and down, the topmost graph on the left is the relationship between the Shannon index and the AVD. To the right, is the relationship between the AVD and the Bray-Curtis dissimilarity. Given that higher AVD would suggest greater diversity, it makes sense that significant ( $p<0.05$ ) positive relationship w/ Shannon and Bray-Curtis. Finally, various nutrient ratios and their relationship with the Shannon diversity index are depicted in the four graphs below, including C:N, C:P, N:P, and the redfield ratio. With the exception of N:P, all ecological stoichiometric relationships had significant positive



	correlation with the Shannon diversity index.
<b>VOCAB: (w/definition)</b>	<p><b>Alpha diveristy index</b> - The mean diversity of species at a local scale. It is the mean species diversity per subunit of the total number of species.</p> <p><b>Beta diveristy index</b> - The ratio of diversity at regional versus local scale.</p> <p><b>Bray-Curtis Dissimilarities</b> - A special statistical tool used in ecology to determine the differences in composition between two specific sites.</p> <p><b>Shannon Index</b> - Another way to measure the diversity of species present in an ecosystem. It is different from the alpha/beta indices in that it is adaptable to multiple scales both ecologically and taxonimically</p> <p><b>Average variation degree (AVD)</b> - Average variation from mean abundance of normally distributed taxonomic groups facing different environmental conditions.</p>
<b>Cited references to follow up on</b>	<p>Vallina, S. M., Cermeno, P., Dutkiewicz, S., Loreau, M., &amp; Montoya, J. M. (2017). Phytoplankton functional diversity increases ecosystem productivity and stability. <i>Ecological Modelling</i>, 361, 184–196. <a href="https://doi.org/10.1016/j.ecolmodel.2017.06.020">https://doi.org/10.1016/j.ecolmodel.2017.06.020</a></p> <p>McCann, K. S. (2000). The diversity-stability debate. <i>Nature</i>, 405, 228– 233. <a href="https://doi.org/10.1038/35012234">https://doi.org/10.1038/35012234</a></p> <p>Zhou, Y., Yang, X., Wang, Y., Li, F., Wang, J., &amp; Tan, L. (2021). Exogenous nutrient inputs restructure phytoplankton community and ecological stoichiometry of eastern Indian Ocean. <i>Ecological Indicators</i>, 127, 107801. <a href="https://doi.org/10.1016/j.ecolind.2021.107801">https://doi.org/10.1016/j.ecolind.2021.107801</a></p> <p>Han, A., Dai, M., Kao, S.-J., Gan, J., Li, Q., Wang, L., Zhai, W., &amp; Wang, L. (2012). Nutrient dynamics and biological consumption in a large continental shelf system under the influence of both a river plume and coastal upwelling. <i>Limnology and Oceanography</i>, 57, 486–502. <a href="https://doi.org/10.4319/lo.2012.57.2.0486">https://doi.org/10.4319/lo.2012.57.2.0486</a></p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. What inter-parameter relationships exist when it comes to the impact exerted upon phytoplankton community composition, structure, and diversity?</li> <li>2. How does light impact nutrient stoichiometry in water body systems?</li> <li>3. Given an increase in phytoplankton biomass, does model accuracy of diversity as well as other attributes decrease?</li> <li>4. How can a time series of phytoplankton community structure and diversity be developed whilst taking into account environmental parameters?</li> </ol>

## NOTES:

## Abstract:

- Aim is to understand the exact relationship between nutrient stoichiometry and the tax. diversity of phytoplankton, specifically in sub-tropical bays
- Phenology observed and community comp in Beibu gulf of South China Sea observed.
- Various families/groups varied phenologically, use of the alpha diveristy index.
- Higher diversity = stabler community (Bray-Curtis Dissimilarities, Shannon Index)

- Redfield Ratio did not impact community structure or diversity, rather, nitrates, phosphorus and temperature did (providing new/unique insight/perspective)

#### Introduction:

- Diversity in phytoplankton helps maintain ecosystem stability, with multiple species present to perform cycling and other necessary functions.
  - Biodiversity = stability
- Many parameters impact their community structure
- Specifically highlights water temp and nutrients
- Ecological change → 2 types of deterministic and stochastic processes in aquatic ecosystems.
- Stability is measured using average variation degree (AVD) (lower value = stability ^)
- Although measured by diversity, article identifies importance in measuring correlation of stability with diversity.
- There is also lack of knowledge on ecological stoich on phytoplankton community structure and diversity
- To analyze these factors surrounding ecological stoich and stability/structure at the spatio-temporally, water samples during four seasons from the Beibu Gulf were analyzed with the ends of:
  - Phenological trends in community structure
  - Identify significant parameters impacting community structure + relationships

#### Methodology:

- Specific latitudes/longitudes where sampling of the Beibu gulf occurred
- Surface seawater data collected 0.5 meter below surface on specific dates from summer 2017 to spring 2018
- n = 100
- Temp, salinity and pH were measured, and filtration systems for DNA were used.
- Various spectrophotometric and colorimetric methods were employed to get data on nitrites, nitrates, phosphates, chl a, total organic C, COD, DO, total dissolved N and P, ammonium, kjedahl N, nutrient stoich ratios
- Specific laboratory procedures and tools used to extract, amplify, and sequence DNA. the sequences of all of the samples collected, were, at the end stored in a database
- 1 - (n/N) (n= specific species size, N = total sample size) was performed (Good's coverage)
- Various R packages
- ANOSIM (analysis of similarity), PERMANOVA and ANOVA all used.
- Spearman's rank method used to to perform correlation analysis
- Alpha diversity calculated using various indices
- Principal *coordinate* analysis (PCoA), AVD, and Bray-Curtis all used as well
- Alpha < 0.05 or <0.01

#### Results:

- See detailed figure analysis above; article material does not offer anything beyond what has been discussed.

#### Conclusions:

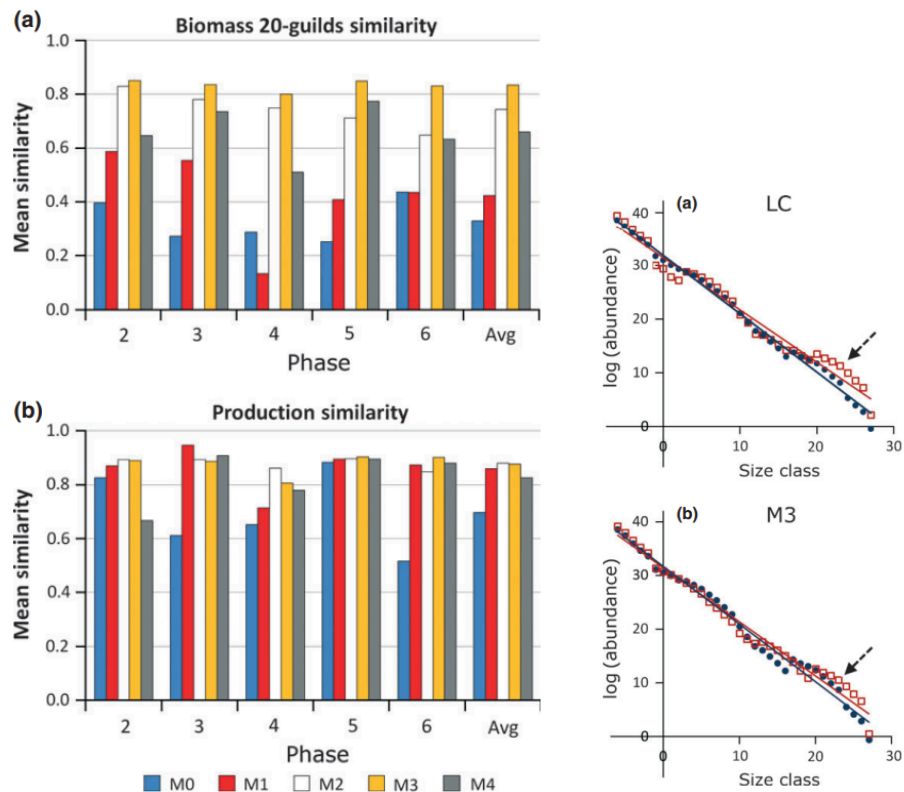
- Low temps arise in the spring waters, hence the drop in diversity observed
- Novelty of the spatio-temporal scale

- These nutrients are significant for the growth and thriving of phytoplankton populations and ecosystem; knowing their values are important
- Bacillariophyceae in winter/spring; Coscinodiscophyceae in summer/autumn;
- Nutrient stoich impacts taxonomic diversity, however more investigation into the underlying factors of these relationships is needed to develop a fuller understanding of what is going on.

## Article #19 Notes: Mechanistic theory and modelling of complex food-web dynamics in Lake Constance

<b>Source Title</b>	Mechanistic theory and modelling of complex food-web dynamics in Lake Constance
<b>Source citation (APA Format)</b>	Boit, A., Martinez, N. D., Williams, R. J., & Gaedke, U. (2012). Mechanistic theory and modelling of complex food-web dynamics in Lake Constance. <i>Ecology Letters</i> , 15(6), 594–602. <a href="https://doi.org/10.1111/j.1461-0248.2012.01777.x">https://doi.org/10.1111/j.1461-0248.2012.01777.x</a>
<b>Original URL</b>	<a href="https://onlinelibrary.wiley.com/doi/10.1111/j.1461-0248.2012.01777.x">https://onlinelibrary.wiley.com/doi/10.1111/j.1461-0248.2012.01777.x</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	Allometric Trophic Network model, community ecology, food web, multi-trophic dynamics, seasonal plankton succession
<b>#Tags</b>	<a href="#">Another, More Detailed Example of Computational Food Web Construction</a>
<b>Summary of key points + notes (include methodology)</b>	The aim of this study is to establish a computational food web in conjunction with a times series to model the ecosystem of Lake Constance. Multiple groups, ranging beyond phytoplankton to includes fish, zooplankton and other organisms, were incorporated to create a food web with 24 guilds and 107 connections. A series of differential equations are used as part of the model. Several iterations of the model, M0 to M4 are developed, each level increasing in complexity and/or refinement. A time series equivalent to eight months is simulated. It is found that M3 is the most accurate model when it comes to predicting attributes such as net production and biomass. This has established a strong starting point for complex computational food webs. Future steps include using more environmental parameters, predicting more ecological characteristics, and projecting on a larger scale.
<b>Research Question/Problem/Need</b>	Using Lake Constance as a case study, how can a neural network of a food web that has an accurate time series be produced, and how the methodology therein be made applicable to multiple situations?

## Important Figures



The figure on the left is an illustration of the similarity of each model in its progressing complexity (M0 - M4) with the measurements of the biomass and production. There are different phases of each model representing the time series aspect of the simulation. It seems that the similarity is the measurement of both parameters is maximized in M3, when the most ecological relationships and parameters are integrated into the model. This is apparent in that most phases, the yellow bar representing M3 is the highest. It is possible that M4, although more refined in having removed potentially unnecessary links, in the process eliminated information, thereby lowering accuracy.

The figure on the right builds on the congruity between M3 and Lake Constance (LC) itself. The regression plot measures biomass against the size of the phytoplankton themselves. The  $r^2$  values are nearly 1, indicating a strong relationship between the two variables both on site and in simulation. There is immense visual similarity between the location of the plots to the median regression line. This is another indicator of M3's accuracy.

**VOCAB:**  
**(w/definition)**

**Guild** - In ecology, a guild is a group of organisms that exploit the same set of resources.

**Allometric Trophic Network** - A special set of network theory techniques used in ecological modelling that uses simple rules (i.e. metabolic/biomass inputs) to model the resulting dynamics.

**Monomictic** - Bodies of water (typically lake/reservoir) that undergo one annual instance of cycling and stratification. Deep, relatively speaking, and no freezing in the winter.

	<b>Exudation</b> - The expulsion of a fluid material from a pore-like structure.
<b>Cited references to follow up on</b>	<p>Williams, R.J. &amp; Martinez, N.D. (2004). Stabilization of chaotic and non-permanent food-web dynamics. <i>Eur. Phys. J. B</i>, 38, 297–303.</p> <p>Williams, R.J. &amp; Martinez, N.D. (2008). Success and its limits among structural models of complex food webs. <i>J. Anim. Ecol.</i>, 77, 512–519.</p> <p>Tirok, K. &amp; Gaedke, U. (2007b). The effect of irradiance, vertical mixing and temperature on spring phytoplankton dynamics under climate change: long-term observations and model analysis. <i>Oecologia</i>, 150, 625–642.</p> <p>Friedrichs, M.A.M., Dusenberry, J.A., Anderson, L.A., Armstrong, R.A., Chai, F., Christian, J.R. et al. (2007). Assessment of skill and portability in regional marine biogeochemical models: role of multiple planktonic groups. <i>J. Geophys. Res.</i>, 112, 1–22.</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How can a mutli-stage, global-scale food web be accurately developed, while taking into account ecological interactions and environmental conditions?</li> <li>2. Given an increase in primary production, is there a decrease in the accuracy of food web models?</li> <li>3. Does an increase in salinity result in the decrease of exudation from cell membrane in phytoplankton?</li> <li>4. How can historical data at a global be harnessed to produce a long-term time scale of a global food web?</li> </ol>

**NOTES:****Abstract:**

- Having models helps understand the highly important dynamics of consumer resources.
- However, this is inhibited by model inability to produce comprehensive food webs.
- Use of neural nets to model pelagic food web of large, well-known lake (good case study)
- Using initial spring biomass and individual body masses, a neural net representing the food web was constructed and parameterized.
- Various measures of energy transfer were incorporated into this network, increasing accuracy of the representation of ecological dynamics.
- Potential new way to model food webs using network theory.

**Introduction:**

- Observing ecological dynamics is important for understanding energy transfer and other critical aspects of ecosystems.
- W/ regards to ecological modeling, computational models of organisms feeding on one another has been limited due to lack of data.
- This is where network theory analysis of food webs becomes key: ecological energy transfer can be better modeled.
- Allometric Trophic Network uses bodily, metabolic properties, biomass fluctuations given resource availability, consumption, and other factors as inputs; fundamental to structure of the neural net model

- Other models have focused on local hydrodynamics and biogeochemical cycling, but given the scope, are not applicable to ecosystems at a broader level — goal is to est. generality.
- Aim was to produce a time series of a neural network food web, something that has not been too heavily evaluated.
- Phytoplankton are used here more as like a case study.
- Lake Constance in Germany/Switzerland/Liech/Austria was used to model the food web, given that it is well-known
- The dataset used contains phenological trends of the phytoplankton: In early spring, a few fast-growing phytoplankton develop during the clear water phase, being highly edible; then the late spring bloom occurs and the diversity increases at the expense of edibility.
- 5 ATNs were used to model this situation, each time with parameter requirements increasing
- This helps better fit the model to accurately capture phenological and ecological trends

#### Methodology:

- Major seasonal trends Lake Constance: in later winter/early spring, irradiance, vertical mixing and temperature control abundance, whereas in the later spring, flowing stratification, nutrient depletion and food quantity and quality are the controls.
- Plenty of data on relevant parameters has been collected for this region
- 24 guilds and 107 feeding relationships were compressed into 20 guilds with 8 groups.
  - Phytoplankton (Phy), autotrophic picoplankton (APP), heterotrophic bacteria (Bac), heterotrophic nanoflagellates (HNF), ciliates (Cil), rotifers (Rot), herbivorous crustaceans (Herb. Cru), and carnivorous crustaceans (Carn. Cru)
  - Species therein are specified
- Time series based on phenological trends was constructed. Temporal phases employed for each season of the year
- Normalization of the data therein was performed to account for differences in the temporal boundaries of these phases.
- Averages attained through the use of a spline function among all the data from the 10 years (1987-1996 (?)) → allows for the weather events to be ignored thereby not confounding anything, and instead focus on the ecological dynamics
- ATN model is based on differential equations related that can be applied to be n species. However, this study adds in metabolism, phytoplankton exudate, and dead particulate and dissolved organic carbon as detritus
- Equations for the foodweb model (takes into account abundance, biomass, metabolism, etc.):

$$\frac{dB_i}{dt} = r_i B_i G_i(\mathbf{B})(1 - s_i) - \sum_j \frac{\alpha_j \gamma_{ji} B_j F_{ji}(\mathbf{B})}{e_{ji}}; \quad G_i(\mathbf{B}) = 1 - \frac{\sum_{j=\text{producers}} \epsilon_{ij} B_j}{K_s}$$

- Producers (the biomass makes use of logistic growth)

$$\frac{dB_i}{dt} = - \frac{\text{maintenance loss}}{f_m} \alpha_i B_i + f_a \alpha_i B_i \sum_j \gamma_{ij} F_{ij}(\mathbf{B}) - \sum_j \frac{\alpha_j \gamma_{ji} B_j F_{ji}(\mathbf{B})}{e_{ji}} - M(\mathbf{B});$$

$$F_{ij}(\mathbf{B}) = \frac{\omega_{ij} \mathbf{B}_j^{q_{ij}}}{\mathbf{B}_i^{q_{ij}} + \sum_{k=\text{consumers}} d_{kj} p_{ik} \mathbf{B}_k \mathbf{B}_i^{q_{kj}} + \sum_{l=\text{resources}} \omega_{il} \mathbf{B}_l^{q_{il}}}$$

- Consumers <sup>^^^</sup> (the bottom equation is also used to make fish guilds conform with the rest of the nodes, as they nonzero mortality values in the model).

$$\frac{dD}{dt} = \sum_i \sum_j \frac{\alpha_{ij} \gamma_{ij} \mathbf{B}_i F_{ij}(\mathbf{B})}{e_{ij}} \quad \text{egested} \quad (1 - e_{ij}) + \sum_i \text{exudation by producers } i \quad r_i \mathbf{B}_i G_i(\mathbf{B}) s_i$$

$$- \sum_j \frac{\alpha_{ji} \gamma_{ji} \mathbf{B}_j F_{ji}(\mathbf{B})}{e_{ji}}, \quad \text{loss by detritivores } j$$

- Detritus matter <sup>^^^</sup>

- M0-M4, 5 iterations of increasing specificity and decreasing generalizability. Models progressively incorporate more and more parameters specific to Lake Constance
- The modelled time series were recorded over 240 time steps translating to 8 months from March to October
- Bray-Curtis similarity to measure the accuracy of the model with regards to parameters
- Statistical variability in the time series was assessed.

#### Results:

- No extinctions occur
- Elaborations upon the overarching trends observed in the above analyzed figure
  - Since M4 does not account for clear water phase characteristics, the model is inaccurate

#### Discussion (Conclusion):

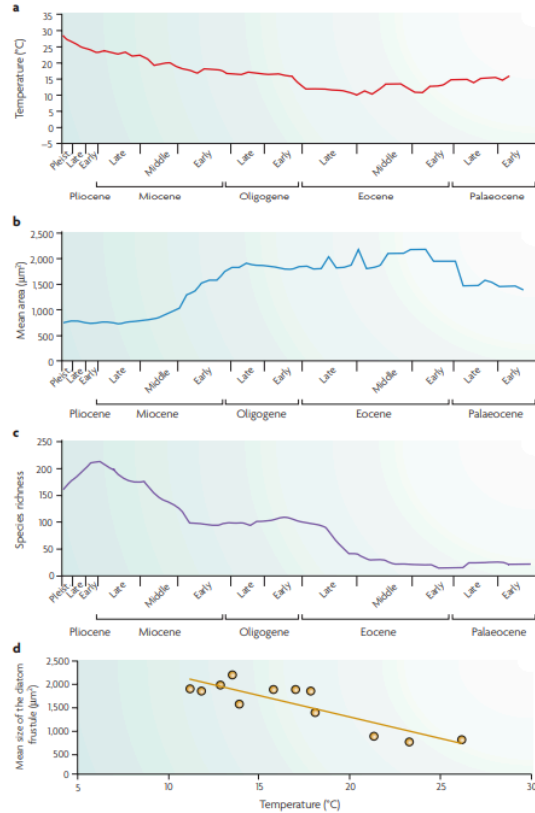
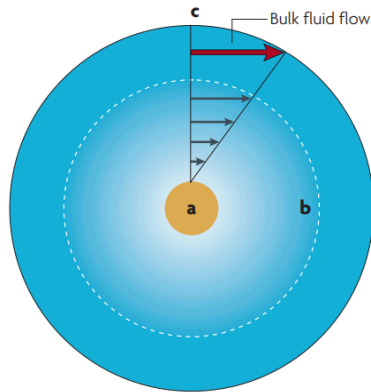
- Computational modeling of food webs has steadily increased in accuracy and scope
- The congruity between the computational model and the field data is novel, especially given the complexity of the model.
- Various adjustments between M0 and M4 needed to be made to perfect model
- Integrating biogeochemistry and ecological stoichiometry, as well as other parameters. Global scale of this model could be developed.
  - Other parameters/eco. factors to consider: prey-predator dynamics, detritus loop, topology, metabolism, abiotic factors, etc.



## Article #20 Notes: Mix and match: how climate selects phytoplankton

<b>Source Title</b>	Mix and match: how climate selects phytoplankton
<b>Source citation (APA Format)</b>	Falkowski, P. G., & Oliver, M. J. (2007). Mix and match: how climate selects phytoplankton. <i>Nature Reviews Microbiology</i> , 5(10), 813–819. <a href="https://doi.org/10.1038/nrmicro1751">https://doi.org/10.1038/nrmicro1751</a>
<b>Original URL</b>	<a href="https://www.nature.com/articles/nrmicro1751">https://www.nature.com/articles/nrmicro1751</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided for this article.
<b>#Tags</b>	<a href="#">An Example Of Phytoplankton-Climate Relations</a>
<b>Summary of key points + notes (include methodology)</b>	The aim of this paper is to use historical trends to project future trends in phytoplankton dynamics, particularly habitat location and taxonomic composition. A line of reasoning is established by reviewing hydrodynamic principles as well as the fossil record. It is shown that these attributes in phytoplankton are regulated by vertical and horizontal nutrient cycling, which are in turn regulated by climate through varying levels of stratification. It is found that these patterns, when combined with the historical evidence, can be used to predict modern day phytoplankton taxonomic composition and habitat. However, limitations include the lack of integration of prokaryotic phytoplankton. For this end, DNA evidence is required.
<b>Research Question/Problem/ Need</b>	How can historical models of climate-phytoplankton relationships be developed to use for future projection, and what scientific theories are needed to establish such relationships?

**Important Figures**



The figure on the left is an illustration of an ideal nutrient concentration and flow fluid surrounding a cell. Flow near the cell surface decreases from bulk-fluid velocity to near zero at the cell surface. Uptake by the cell depletes the nutrient concentration around the cell. This necessitates a strong nutrient gradient within the boundary layer. The dashed line represents two different layers: the layer closer to the cell has slower movement, making diffusion even more challenging.

On the right, many correlative relationships from the hisotrical data are presented. The topmost graph is the global average temperature over the past few geological ages. Although the patterns seems to show a decline that plateaus, there is a noticeable upward tick right at the end, indicating current global warming. The second illustrates marine diatom shell area. During the Eocene, the area peaked, before declining somewhat to its current day size. Species richness over time has steeply declined. This is an intriguing finding to consider. The bottommost graph illustrates a negative correlation between temperature and marine diatom size.

**VOCAB: (w/definition)**

**Bulk fluid** - The main direction in which the fluid travels, and it also references the materials it holds therein.

**The Kolmogorov cascade** - A measure of length and scale of oceanic eddies as a means of measuring circulation

**The Reynolds number** - A metric the measures whether a fluid is influenced by inertial or viscus forces.

	<b>Riverine</b> - Nutrients that are discharged by rivers/streams.
<b>Cited references to follow up on</b>	<p>Grover, J. P. Dynamics of competition among microalgae in variable environments: experimental tests of alternative models. <i>OIKOS</i> 62, 231–243 (1991).</p> <p>Tozzi, S. Historical climate change and ocean turbulence as selective agents for two key phytoplankton functional groups. <i>Mar. Ecol. Prog. Ser.</i> 274, 123–132 (2004).</p> <p>Connolly, J. et al. Correlated evolution of cell volume and genome size in diatoms (Bacillariophyceae). <i>J. Phycol.</i> (in the press).</p> <p>Katz, M. E., Finkel, Z. V., Grzebyk, D., Knoll, A. H. &amp; Falkowski, P. G. Evolutionary trajectories and biogeochemical impacts of marine eukaryotic phytoplankton. <i>Annu. Rev. Ecol. Evol. Syst.</i> 35, 523–556 (2004).</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. Do eukaryotic phytoplankton offer significantly more nutrients to an ecosystem than their prokaryotic counterparts?</li> <li>2. In the broader picture of energy transfer, how strong is the impact of varying absorption rates in phytoplankton?</li> <li>3. Are there any situations analogous in history to the current conditions of global warming that can be fully modeled?</li> <li>4. How has continental drift impact ocean conditions, and in turn, phytoplankton dynamics?</li> </ol>

**NOTES:****Abstract:**

- Aim is to understand poorly understand impact of climate on microorganisms, specifically phytoplankton
- Climate-phytoplankton relationship determined using competition theory, fossil record.
- Thermal influences of climate influence mixing, which determines nutrient fluxes that influence tax comp and cell size
- Understanding the connections between the above stated processes shall allow for better basis of predictions on how phytoplankton will change due to global warming.

**Introduction:**

- Historically, metabolic activity has been able to adapt to sudden changes, though abundance has varied a lot.
- Using prognostic models on phytoplankton dynamics is difficult with a lack of historical data. Hence, historical models would be useful.
- Eukaryotic phytoplankton have a comparatively voluminous fossil record for these ends. Although accuracy cannot be ensured, a good frame of reference is offered.
- Climate → ocean mixing → phytoplankton population dynamics
- Being able to characterize this will not be fully complete until the prokaryotic phytoplankton too are incorporated have enough evidence to base this theory off of.
- Focus is on dinos, diats, and coccos.

**Methodology:**

- The article provides background the basics of ocean layer mixing and ocean structure
  - Pelagic/Benthic ocean varies in terms of nutrient concentration
  - There are two types of nutrient cyclings that result: isopycnal, which is the mixing coming in from the top with the bottom layer, and diapycnal mixing, which is mixing that comes from the ocean floor
- Differential equation for vertical density gradient established.
- Article discusses the connection between nutrient kinetics, uptakes, and mixing, providing key relationships in the form of certain equations:

$$\text{growth} = \mu_{\infty} \left(1 - \frac{Q_{min}}{Q}\right)$$

$$\text{uptake} = V_{max} \frac{R}{K + R}$$

$$R^* = \frac{K\mu_{\infty}Q_{min}m}{V_{max}(\mu_{\infty} - m) - \mu_{\infty}Q_{min}m}$$

- Focus is placed on the various biochemical and hydrodynamical parameters that inhibit diffusion of nutrients into the cell membrane
- With these premises established, the fossil record of phytoplankton is now investigated
  - Through historical analysis, it is determined that there is a direct impact that the climate exerts on ocean conditions
- In turn, the article hypothesizes that the climate impact the phytoplankton by controlling their habitat location (i.e. selecting it for them) on the basis of the exertions on nutrient availability.

#### Results:

- See Analysis of Figure 5 above; provides significant correlations for the findings of the study.
- Article asserts that it is possible to use the principles of nutrient circulation established as well as the historical data to make predictions about future phytoplankton habitat location and thereby the tax comp.
- Makes some preliminary predictions on this basis, such as that coccolithophorids will come to dominate, as they are well-adapted to low nutrient environments which stratification increasing the presence of.
- Limitations: This model can only be applied to eukaryotic phytoplankton; no fossil evidence of prokaryotes has been provided → calls for the use of DNA evidence

## Article #21 Notes: Causal networks of phytoplankton diversity and biomass are modulated by environmental context

<b>Source Title</b>	Causal networks of phytoplankton diversity and biomass are modulated by environmental context
<b>Source citation (APA Format)</b>	Chang, C.-W., Miki, T., Ye, H., Souissi, S., Adrian, R., Anneville, O., Agasild, H., Ban, S., Be'eri-Shlevin, Y., Chiang, Y.-R., Feuchtmayr, H., Gal, G., Ichise, S., Kagami, M., Kumagai, M., Liu, X., Matsuzaki, S.-I. S., Manca, M. M., Nôges, P., & Piscia, R. (2022). Causal networks of phytoplankton diversity and biomass are modulated by environmental context. <i>Nature Communications</i> , 13(1), 1140. <a href="https://doi.org/10.1038/s41467-022-28761-3">https://doi.org/10.1038/s41467-022-28761-3</a>
<b>Original URL</b>	<a href="https://www.nature.com/articles/s41467-022-28761-3#MOESM1">https://www.nature.com/articles/s41467-022-28761-3#MOESM1</a>
<b>Source type</b>	Journal Article
<b>Keywords</b>	No keywords are provided.
<b>#Tags</b>	<a href="#">Instance of using chlorophyll as an indicator for phytoplankton dynamics</a> <a href="#">Potential time series model analysis tool</a>
<b>Summary of key points + notes (include methodology)</b>	This study takes a holistic approach to identifying the influencers of driving parameters of phytoplankton dynamics across different ecosystems conditions. An exogenous variable, temperature is linked to the phytoplankton dynamics of focus, biodiversity (based off species index), and biomass (approximated from chl a concentrations). These two characteristics were casually linked to each other as well as to nitrate and phosphate concentrations, forming pairwise and triangular networks of casual relationships whose strengths were evaluated across 19 diverse aquatic ecosystems in the Northern Hemisphere. Using CCM, causality among parameters between the different ecosystems was calculated, with nonlinear time series models having been created for each parameter in each ecosystem. Using redundancy analysis, various conclusions about the nature of inter-parameter relationships were successfully reached, proving the viability of this method. For example, it is predicted that since warmer water temperatures increase the influence of nitrates, with warming ocean conditions, it is predicted that nitrates will become a prevalent driving factor (a myriad of conclusions like this were reached). Further data implementation is needed to validate statistical analysis and model validity and allow for an even more holistic perspective.
<b>Research Question/Problem/ Need</b>	How can the nature of causal networks among environmental parameters be revealed, how does that vary across differen ecosystem conditions, and what ramifications do such attributes suggest?

Important Figures

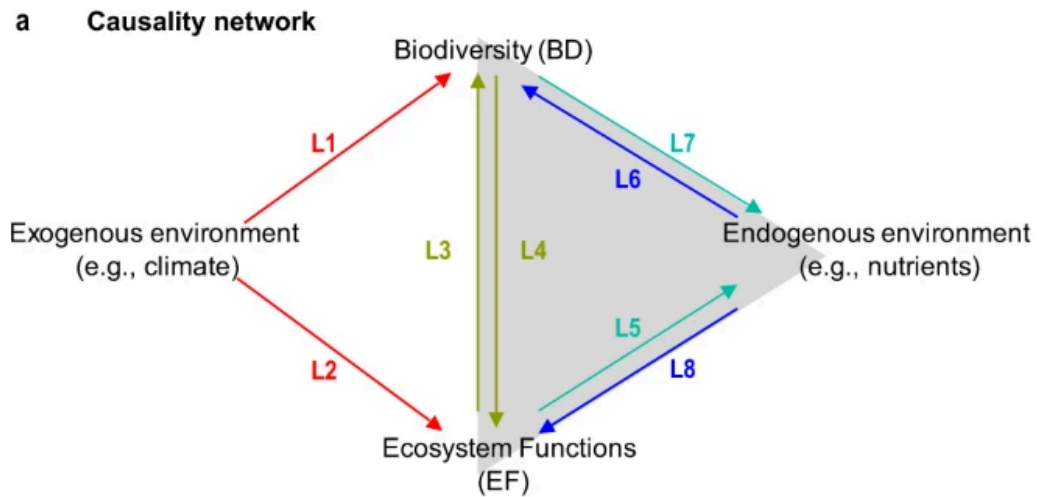


Figure 1a illustrates the various causal networks among broad, crucial ecological metrics. Biodiversity and ecosystems functions, as posited by the theory of feedback, have a mutually casual relationship (L3, L4). Exogenous variable, including climatic factors, tend to exert unidirectional impacts on BDEF (which I might disagree with, as phytoplankton dynamics changing would mean greater ramifications for some aspects of climate). Meanwhile, endogenous factors, i.e., typical environmental parameters that you've tended to focus on, have mutually reinforcing relationship with BDEF.

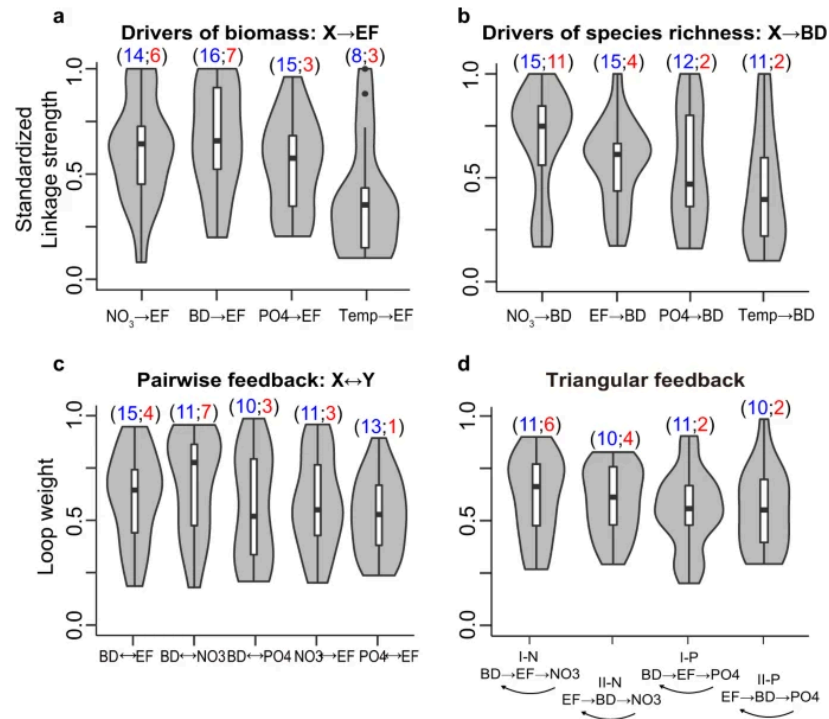
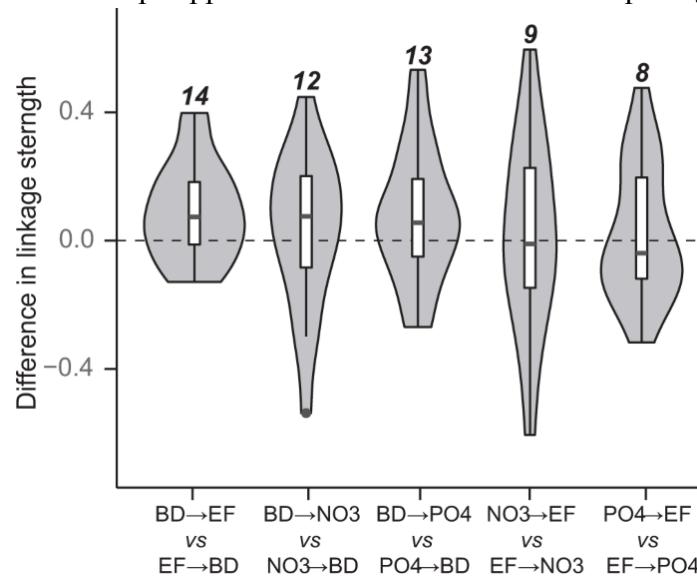


Figure 2 illustrates the various linkage strengths of parameters to ecological qualifiers, namely biomass (EF) and species richness (BD). From top left to bottom

right, we see that the linkage strength of parameters to EF is around 0.7, with the exception of temperature, which has a weaker strength, standing at about 0.4. This is complemented by corresponding distribution of the boxplot and histogram. This means that micronutrients and biodiversity are key to determining biomass. Similar means of analysis reveal that nitrate has the greatest influence over biodiversity, followed by biomass, and far behind phosphate and temperature. The pairwise loop weight (as this is now multi-directional), between biodiversity and nitrates as well as biodiversity and biomass are much greater than any other interactions. All triangular relationships appear to have similar levels of loop weight, between 0.6-0.7.



This diagram (figure 3) compares linkage strength of pairwise, looking specifically at which direction holds a stronger link. A certain direction between pairs is chosen, and the number on top represents the number of ecosystems (of the 19) where that linkage was stronger than its opposite. Based off analysis on the range and distribution of the violin plots, and the numbers denoting the amount of ecosystems where the specific pair is stronger than its opposite, it is clear that biodiversity is more impactful towards other parameters than the other way around. Meanwhile, phosphate and nitrate seem to have a stronger link with biomass than the other way around. Meaning, the nutrients more strongly influence biomass than biomass does the nutrients.

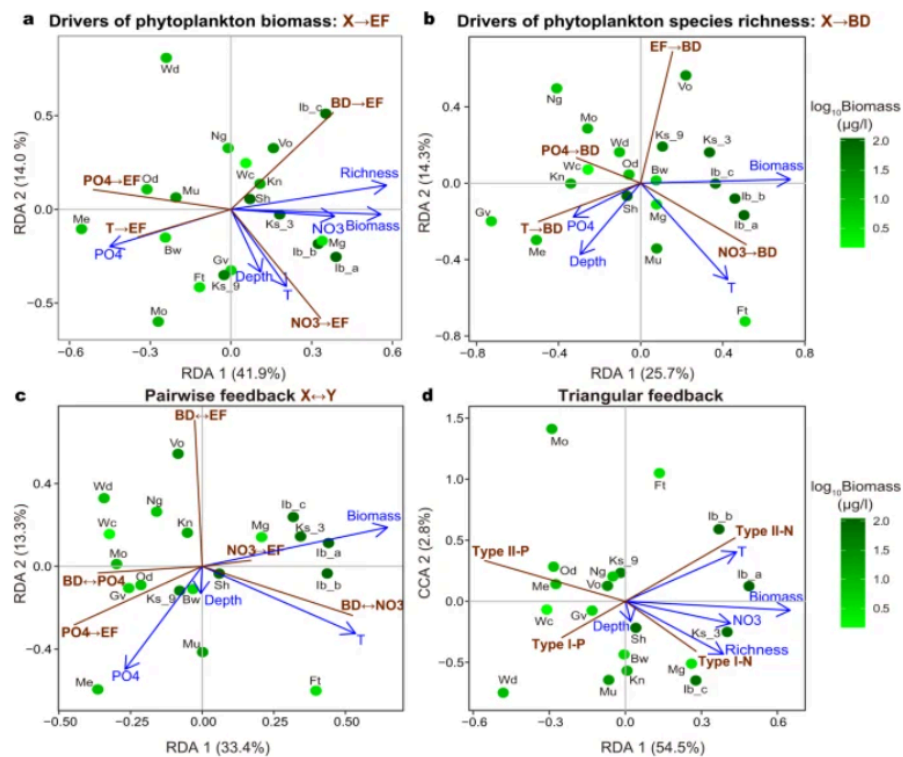


Figure 4 illustrates redundancy analysis plots (RDA). Using the principles of RDA (similar to PCA), we can arrive at conclusions for environmental context. The blue vectors represent the raw parametric values, and the brown ones represent the directional networks being observed. Antiparallel vectors are negatively correlated, whereas adjacent ones are positively correlated.

For 4a, biodiversity most strongly impacts biomass in low-phosphate environments, and in moderate depths, temperature, and NO<sub>3</sub> values. Meanwhile, PO<sub>4</sub> has the strongest causal links with biomass in lower depths (meaning shallower), temperatures, diversity and NO<sub>3</sub>. NO<sub>3</sub> is most strongly linked w/ EF in higher amounts of these parameters. Temperature and phosphate have a near synergistic impact on phytoplankton biomass. (insight below)

For 4b, biomass most strongly impacts biomass in shallow, cooler, oligotrophic conditions. The opposite is true for the causal link between temperature and biodiversity. Higher temperatures allow for stronger nitrate→diversity links (reinforcing conclusion from 4a). Meanwhile, PO<sub>4</sub>→diversity is strongest when temperature and biomass are lower.

For 4c, BD ⇌ NO<sub>3</sub> is strongest under warmer temperatures. BD ⇌ EF is strongest when depth, PO<sub>4</sub> and temperature are lower. For BD ⇌ PO<sub>4</sub> and EF ⇌ PO<sub>4</sub>, higher phosphate levels (unsurprisingly) facilitate strength between these causal relationships, combined with low biomass. By contrast, NO<sub>3</sub> ⇌ PO<sub>4</sub> rely on lower phosphate and greater biomass for stronger causal relationships.

Finally, for 4d, greater depth NO<sub>3</sub>, biomass and biodiversity facilitate BD → EF → N → ... strength, whereas the opposite is true for the EF → BD → P → ... cycle.



	Lying almost as reference angles in the other quadrants are $EF \rightarrow BD \rightarrow N \rightarrow \dots$ , which is strongest under warmer temperatures, and $BD \rightarrow EF \rightarrow P \rightarrow \dots$ , strongest under cooler temperatures.
<b>VOCAB: (w/definition)</b>	<p><b>Feedback</b> - In the context of ecology, the co-interaction between evolution and the environment whereby environmental conditions form the trajectory of evolutionary patterns which in turn result in environmental conditions, and this continues the cycle of mutual interaction.</p> <p><b>Oligotrophic</b> - The opposite of eutrophic. Whereas eutrophic conditions and eutrophication involves excessive nutrients that form algal blooms and anoxic conditions, oligotrophic systems have a dearth of plant nutrients and an abundance of oxygen.</p> <p><b>Convergent Cross-Mapping</b> - Method used to identify cross-system variable relationships and causality between different variables, Causality analysis is based on Takens' theorem for dynamical systems which infers the causal relationship among variables from their empirical time series.</p> <p><b>Kendall's <math>\tau</math> test</b> - A nonparametric approach to using statistical test results to rank categorical variables. (delta-rho Z likely a type of follow up test, similar to Dunn's for Kruskal-Wallis, Tukey for ANOVA)</p>
<b>Cited references to follow up on</b>	<p>Mueller, K. E., Tilman, D., Fornara, D. A. &amp; Hobbie, S. E. Root depth distribution and the diversity–productivity relationship in a long-term grassland experiment. <i>Ecology</i> 94, 787–793 (2013).</p> <p>Pikitch, E. K. et al. Ecosystem-based fishery management. <i>Science</i> 305, 346–347 (2004).</p> <p>Sugihara, G. et al. Detecting causality in complex ecosystems. <i>Science</i> 338, 496–500 (2012).</p> <p>Oksanen, J. et al. <i>vegan: Community ecology package</i>. R package version 2.5-7. <a href="https://CRAN.R-project.org/package=vegan">https://CRAN.R-project.org/package=vegan</a> (2020).</p> <p>Ye, H. et al. <i>rEDM: Applications of empirical dynamic modeling (EDM) from time series</i>. R package version 1.2.3. <a href="https://CRAN.R-project.org/package=rEDM">https://CRAN.R-project.org/package=rEDM</a> (2020).</p>
<b>Follow up Questions</b>	<ol style="list-style-type: none"> <li>1. How would other parameters (oxygen, salinity, silicates, etc.) impact the nature of the causal networks observed?</li> <li>2. How can the implications of causal networks among driving parameters across multiple ecosystems be linked to neural networks for measuring trophic energy transfer in the food web?</li> <li>3. How do redundancy analysis and principal component analysis compare in terms of the efficacy of identifying the ecological context of the strength of causal relationships?</li> <li>4. How can climate models, name the CMIP, integrate inter-parameter analyses</li> </ol>

	<p>in the form of networks such as the ones used in this study?</p> <p>5. Can this empirical methodology be applied to other scenarios?</p>
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## NOTES

### Abstract:

- It is difficult to assess driving factors of ecological biodiversity and other crucial factors among different ecosystems due to the numerous circumstances surrounding them
- Meaning, unifactor experiments have highly inapplicable results
- 19 ecosystems within the midlatitudes of the Northern Hemisphere were investigated.
- As diversity and oligotrophic conditions increase, biodiversity factors supplant environmental ones as driving factors of biomass (make sense)
- Warmer, more productive ecosystems: Nitrate-mediation; Colder, less productive ecosystems: Phosphate-mediation
- Analyzing ecosystems through the lens of networks and network theory is crucial for future management of ecosystems

### Introduction:

- The idea of **feedback** has been important for global-scale analysis of ecosystems. See definition above, but in network theory, this basically involves nodes and the causal interactions that come from it
- Looking at feedbacks has been suggested as a way of understanding the interaction between biodiversity/ecosystem function (BDEF) regulate nature
- However, there are also environmental parameters to consider.
- Herein lies conceptual networks of the causal interactions between these various ecological metrics (figure 1 analysis above)
- There exists a need to quantify these networks
- Once again, experiments fail to provide a full, applicable picture due to unidirectionality omitting the analysis of the impact of all other factors
- Regression approaches assume consistency among factor relationships, failing to be fluid in accounting for different time periods and environmental conditions, which are dynamic (exactly what I found as a limitation in my current project progress)
- Experimentally constructed ecosystems have diversity as the main driver, whereas natural systems are more complex in determining these factors
- Quantification helps make the picture clearer in this regard; it can also predict potential outside stressors on ecosystem dynamics
- The article asserts that it is helpful to analyze the lack of unidirectionality of the triangular networks in addition to that of the pairwise functions. That way, systems as a whole are analyzed rather than just individual relationships
- This study focuses on the importance of BDEF for marine ecosystems. This is novel as most knowledge of these broad ecological qualifiers is based on results from terrestrial ecosystems
- This is specifically problematic considering the role of phytoplankton as >50% of primary production source; phytoplankton have only received marginal attention.

- Using nonlinear time series models and convergent cross-mapping (CCM, see definition above), an analysis of the networks within and between 19 sites among 16 globally distributed systems was performed.
- Biodiversity characterized using species richness indices; ecosystem function → biomass, given from approximation from chlorophyll a; NO<sub>3</sub>, PO<sub>4</sub>, water temperature were also used.
- Following four questions were analyzed:
  - 1. When does phytoplankton diversity outweigh the environmental parameters as a driver of ecosystem functioning?
  - 2. What is the strongest causal determinant for species diversity?
  - 3. What ramifications for broad ecological qualifiers does a change in phytoplankton diversity pose?
  - 4. Do there exist macroecological factors in assessing the nature of networks between these broad ecological qualifiers
- The following four things were evaluated:
  - 1. Drivers of phytoplankton biomass
  - 2. Drivers of phytoplankton diversity
  - 3. Analysis of links between drivers of biodiversity
  - 4. How the strengths of the networks studied in 1-3 vary given environmental conditions
- Results indicate that in more diverse, oligotrophic systems, diversity plays a bigger role in biomass. In most systems, diversity is driven by nutrient concentrations.
  - Warmer, more productive ecosystems: network between nitrate-diversity-biomass
  - Colder, less productive ecosystems: network btwn phosphate-diveristy-biomass

#### Methodology:

- 16 Systems, 19 monitoring sites across the northern hemisphere (japan, europe, usa mainly)
- 5 Variables
  - Species richness (# of dif. species)
  - Chl a, common proxy for BDEF
  - Phopshate
  - Nitrate
  - Water Temp
- As well as: water column stability, irradiance, and zooplankton abundance, which also impact phytoplankton populations
- Impact of herbavorous zooplankton species, including cladocerans, copepods and crusteceans was taken
- Data had to be clean to fit the equal time-series intervals of monthly data, but CCM data was still validly carried out
- 5554 datapoints for each variable for each of the 19 sights.
- Long-term linear trend removed to maintain proper trend characterization of function. Seasonality accounted for using mean and standard deviation values.
- Biomass, diversity, and other indicators and networks had their relationships evaluated with CCM

- See def above + can determine the degree to which time “causes”, or rather, predicts, the level of the causal variable, as well as inter-time series causality (not just at variable level)
- Various means taken to minimize the pitfalls of CCM strategy, such as deseasonalization to account for false positive identification
- Interaction among environmental parameters tends to be weak to moderate, if at all ([here](#))
- The multiscale-analysis’ validity is limited, due to data dearth and poor structure (too many leading/lagging 0’s, inconsistent data implementation into time series for CCM)
- Using lagged embedding, which looks at the retrospective variable and network measurements as the time series forms, confounding variables and their effects may be identified.
- Various time series operations involving the functions of various variable pairs are performed to allow for this investigation of confounding variables
- Though this dampens the strength of quantitative analysis, it allows for causality to easily be inferred. Correlations and relationships are able to dynamically shift on the temporal scale.
- Kendall’s  $\tau$  test and Fisher’s  $\Delta\rho Z$  test used to validate CCM use
- CCMs measure the strength of causality between two variables, as well as the impact of external factors.
- The correlation calculated were all standardized among one another.
  - Linkage strength (calculated using CCM) → Standardized linkage strength
  - Minmax
- Neutel’s loop weight taken by using geometric mean of linkage strengths. Bootstrapped intervals from 500 total sampling instances.
- Redundancy Analysis (similar to PCA) was performed for: drivers of biomass and diversity, as well as for the triangular neural nets
  - Not for causation, but rather for driver identification
  - 10,000 samples pseudo-bootstrapping method
- R

## Results and Discussion

- Phytoplankton diversity is a major controlling factor biomass, despite the 19 very different ecosystems (makes sense, intuitively)
- Nitrate concentration had a similar level of significance for this factor as well.
- See figure 2 analysis.
- On the other side of the coin, while biomass drives biodiversity, nitrate is the stronger driver.
- Nitrogen is a significant determinant of both biomass and biodiversity
- Using Shannon Diveristy index as an alternative measurement, same results were found, although biomass was shown to be more important.
- Therefore, as global warming modifies nutrient conditions, it will be important to watch how phytoplankton diversity and biomass change as a result.
- However, phosphate is also another significant nutrient to consider, as its interactions with nitrogen are not fully clear, and require further investigation
- When also looking at irradiance, column stability, and zooplankton, really the only significant parameter identified was the feeding of crustacean zooplankton.

- Figure 3 analysis
- Strengths of the  $BD \rightleftharpoons NO_3$  were highly variable, though having the highest median linkage strength, whereas  $BD \rightleftharpoons EF$  was less variable and had greater significance among the 19 ecosystems studied (15 vs. 11).
- We see that (1) environmental context and conditions formulate whether nutrients, ecological factors, or other parameters influence phytoplankton dynamics, and (2) that BD is a common factor here. Thus, bidirectional relationship networks also ought to be considered. (i.e.  $EF \leftarrow BD \rightarrow NO_3$ ).
- Looking at the triangular feedbacks between nutrient to biodiversity to biomass to..., when comparing phosphates and nitrates, and inputting them as “nutrients,” within the described feedback loop (as seen below), nitrate tended to have stronger links to these ecological characteristics more so than P, though not to a statistically significant degree.
  - Type I-N:  $BD \rightarrow EF \rightarrow N \rightarrow$
  - Type II-N:  $EF \rightarrow BD \rightarrow N \rightarrow$
  - Type I-P:  $BD \rightarrow EF \rightarrow P \rightarrow$
  - Type II-P:  $EF \rightarrow BD \rightarrow P \rightarrow$
- More of the lakes studied were eutrophic, and therefore less nitrogen heavy, thus making nitrogen concentrations a stronger focal point for ecosystem structure (of phytoplankton)
- Between Type I and Type II, there were no significant differences in linkage strength. Meaning, BD could impact biomass directly as well as indirectly through nutrient concentrations, and a similar idea for EF and N. (makes sense – think about a triangle)
- How environmental context impacts which variables have stronger links and greater influence over phytoplankton can be discerned using RDA chart and analyzing vectors. Below notes provide ramifications, and figure 4 analysis above specifies environmental contexts.
- Synergistic impact on biomass and temperature and eutrophic conditions matches findings in past literature. However, based off RDA chart, diversity is negatively correlated with these two variables, as is the case when  $BD \rightarrow$  biomass is greater. Meaning, warming and eutrophic conditions, which are becoming increasing problems for biodiversity, however, biodiversity is the key to combating these problems (potential pursuit for policy)
- Shallower systems are comparatively homogenous, which allow for species interactions to be more influential (given lower temp and  $PO_4$  and shallow  $\rightarrow$  stronger  $EF > BD$ ). Additionally, when temperatures were warmer,  $NO_3 > BD$  was stronger, indicating that drivers of biodiversity were determinants resulting from a temperature gradient.
- Based off the inter-parameter relationships depicting in 4c, we can say that the colder water temperatures allow for stronger  $PO_4$  mediated feedbacks, whereas warmer temperatures allow for stronger  $NO_3 \leftarrow \rightarrow BD$  impacts, which matches overall literature findings that N is more influential when temperature is warmer. Thus, as oceans warm,  $NO_3$  is projected to become very important in determining biodiversity.
- From 4d, the stronger the biodiversity, the stronger the causal cycle is between biomass, biodiversity, and environmental conditions, meaning there is higher response, change, and therefore, stability, when diversity is higher. This contradicts the consensus that biodiversity encourages insensitivity and stability in face of environmental change, thus illustrating the need to clarify the impacts of biodiversity on ecological dynamics

- Causal links of the same parameters but order differently can respond different given inputs of environmental parameters, creating another layer of complexity to consider
- Highlights the needs for holistic computational approach
- Robust to different values sensitivity analysis
- Possible extension: looking at the impacts on taxonomic composition.
- Limitations the feedback networks could have their sign values accessed, meaning whether there is a  $\pm$  is unknown.
- Data availability and number of systems studied could be enlarged, i.e. iteration needed
- Overall, study effectively bridges various elements of ecological analysis and characterization and conditions of ecosystems. Causal networks ought to be analyzed, as they offer an effective approach. Improved understanding and considerable ramifications were achieved.
- HUGE ramifications for project methodology— using RDA to identify *when* parameters are significant, and not just the driving factors, allows for a sort of PCA<sup>2</sup>, wherein the driving parameters *of* the driving parameters can be understood— very powerful tool to potentially integrate into computational model.

## Patent #1 Notes:

APA Citation:

Hiroharu, M. K., Masahiro, Y., & Okamura, O. I. (2022). *Calculation method and calculation device for the abundance of phytoplankton of a specific species, and sign detection method and sign detection device for red tide occurrence by phytoplankton of a specific species.*

[https://patents.google.com/patent/JP7007225B2/en?q=\(phytoplankton\)&oq=phytoplankton](https://patents.google.com/patent/JP7007225B2/en?q=(phytoplankton)&oq=phytoplankton)

Title:

Calculation method and calculation device for the abundance of phytoplankton of a specific species, and sign detection method and sign detection device for red tide occurrence by phytoplankton of a specific species.

URL: [https://patents.google.com/patent/JP7007225B2/en?q=\(phytoplankton\)&oq=phytoplankton](https://patents.google.com/patent/JP7007225B2/en?q=(phytoplankton)&oq=phytoplankton)

Summary/Notes:

The device is aimed at using varying phytoplankton fluorescence properties to identify specific species, and thereby abundance, composition and other properties. It is also designed to check if there is a red tide. A red tide is indicative of certain species of phytoplankton that have been found to be detrimental to economic and environmental well being (harming both fishing industry output and the stability of marine food webs). Hence, red tide identification has been a major priority.

*Major question:* What advantages and disadvantages does this device pose, relative to the use of genomic methods?

- Light calculations: As described, light properties are exploited to identify phytoplankton. More broadly, many of the optical properties of the water are used including absorption and scattering.
- Measurements are then analyzed by the tool. Beforehand, a desired phytoplankton species is chosen, and the light calculations calculate the presence of that species taking into account the species that may be present in the sample
- These properties can also be used to help identify red tides
  - This process involves temporal analysis of light properties in the water.
- Part of the apparatus contains a calculation device that takes into account the red tide measurements and the light properties, providing the gross calculations for the phytoplankton species identification and abundance measurements
- Compared to traditional methods (i.e. typical testing tool kits), this tool is advantageous:
  - Live coverage of water quality
  - Early warning system for red tides
  - Test results are overall faster, more reliable, and accurate.
- Very useful for management purposes, fisheries, health monitoring of the ecosystem, and the like

## Patent #2 Notes:

APA Citation:

Terada, M. (2020). *Phytoplankton growing apparatus and phytoplankton growing method*.  
[https://patents.google.com/patent/WO2020070733A1/en?q=\(phytoplankton\)&oq=phytoplankton](https://patents.google.com/patent/WO2020070733A1/en?q=(phytoplankton)&oq=phytoplankton)

Title:

Phytoplankton growing apparatus and phytoplankton growing method

URL:

[https://patents.google.com/patent/WO2020070733A1/en?q=\(phytoplankton\)&oq=phytoplankton](https://patents.google.com/patent/WO2020070733A1/en?q=(phytoplankton)&oq=phytoplankton)

Summary/Notes:

The patent proposes a circular device acting as a medium for phytoplankton growth. It involves a circulation mechanism so as to stimulate and equilibrate conditions within the contraption. The phytoplankton are pushed upwards via a pumping mechanism where they are subsequently exposed to light. If and when more abundance of phytoplankton is needed, methods such as these are crucial.

Deeper analysis of the structure of the device:

- The circulation apparatus is in fact helpful in allowing the phytoplankton to remain in an environment where they have sufficient light and nutrient exposure, thereby maximizing growth
- This is especially powerful when considering the fact that there is a vertical pump mechanism that sends the phytoplankton upward. At this destination lies the upper cover of the circular contraption, allowing for light exposure.
- Stacked structures, known as flanges are located within the structure, allowing for downward circulation. This completes the cycling of nutrients, allowing for optimal conditions.

Consequently, this device is more efficient than most of its peer horizontal tanks.

- Space efficient, with an internal equilibrium maintained
- The light-circulation mechanism is both novel and increase efficiency of the growth
- Although the model is made to be space efficient, what is more is that it is also able to be scaled up or down, allowing for an additional dimension of flexibility and optimization

There are many areas in which this patent can be applied. Namely, it would be useful for researchers, as they would have any easy way to grow phytoplankton for experimentation. There would be other uses with regards to R&D. The increased harvests could also serve as biofuel, feeding, and other useful ecological and economic purposes.



## Patent #3 Notes:

APA Citation:

Zhao, Y. G., Lishi, N. H., Xiaoya, C. Y., Jingbo, F. L. D., Gan, X. X., Xinxin, T. D., Liu, Z. Y., & Wenqing, J. L. (2016). *Based on phytoplankton identification assay method and the device of discrete three-dimensional fluorescence spectrum.*

[https://patents.google.com/patent/CN103868901B/en?q=\(phytoplankton\)&oq=phytoplankton+](https://patents.google.com/patent/CN103868901B/en?q=(phytoplankton)&oq=phytoplankton+)

Title:

Based on phytoplankton identification assay method and the device of discrete three-dimensional fluorescence spectrum

URL:

[https://patents.google.com/patent/CN103868901B/en?q=\(phytoplankton\)&oq=phytoplankton+](https://patents.google.com/patent/CN103868901B/en?q=(phytoplankton)&oq=phytoplankton+)

Summary/Notes:

This patent, similar to the first one, is aimed at identifying phytoplankton by taking advantage of different species' fluorescence properties. What is novel about this approach its use of a discrete three-dimensional fluorescence spectrum. This provides a convenient gradient by which known phytoplankton irradiance patterns can be compared to. This provides a clean sort of assay process in determining the phytoplankton species. The device, compared to Patent #1, is smaller and easier to operate. Hence, having methods to determine these characteristics quickly and well is very important. That way, not only can the crucial characteristics be measured, but this can be done efficiently.

More details on methodology:

- A device is used to take pictures of and observe the 3D fluorescence patterns of individual cells
- The specification and accuracy in identifications is more accurate than pigment and morphological methods, which have been used hitherto. Relies on less convoluted methods too, with the omission of pigmentation, chromatography and other complex apparatuses.
  - What is more is that it can operate very rapidly.
- Spectrometric methods and laser technology are utilized to gain data from the cytological fluorescence
- From the data attained, identification is performed via database comparison

Additional advantages:

- Its rapid testing is preferable to pigment and morphological methods, which are known to be convoluted and time-consuming
- By being able to identify species, even those that are similar, this model breaches a limitation that previous methods failed to
- The cells are analyzed without being harmed → allows for empirical evidence w/o environmental harm (a win-win situation)
- Versatility applicable to many species and ecosystems

Applications are wide ranging as stated above, but notable areas of application include management of aquatic ecosystems, broad study of phytoplankton dynamics (*very topical for this project*), tracking, predicting, and addressing algal blooms, environmental monitoring, and biofuel and other economic ventures.

**Overall, these patents offer practical solutions to the computational problem being assessed for the project.**