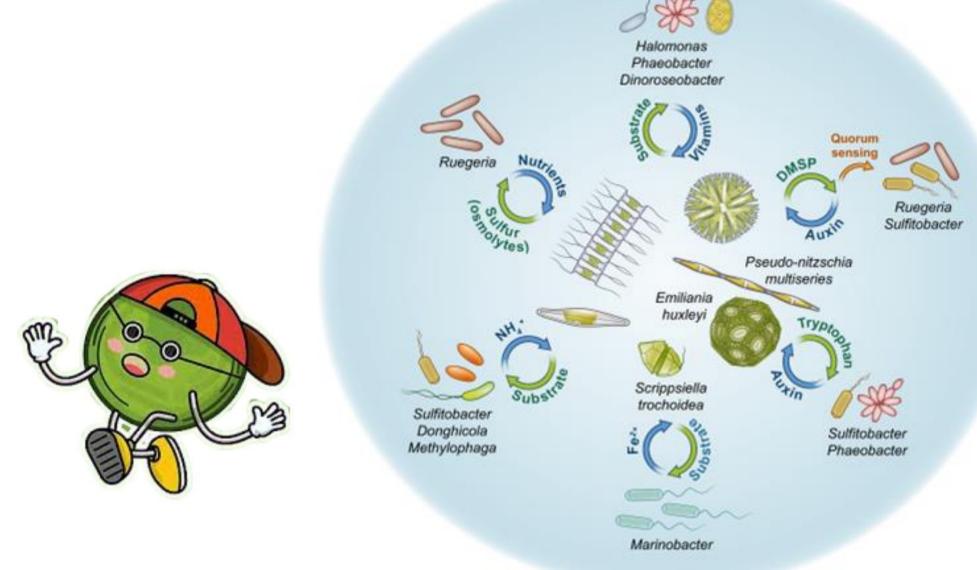
Carbon source impacts algal growth and shapes microbiomes in outdoor ponds

Duke Sass Connections

Introduction

Marine microalgae are essential to the functioning and health of aquatic ecosystems as keystone species and providers of an estimated 50% of atmospheric oxygen among other ecosystem services. Recent interest in large scale microalgae cultivation is driven by their strong potential for carbon capture as well as commercial applications such as biofuels, pharmaceuticals, foods, cosmetics, and even plastics. Large scale algae cultivation is often done in large, open raceway ponds, and efforts to optimize this process have led to developments in the growth media composition, bioreactor design, strain of algae and conditions of light and gas exchange.



However, these microalgae do not grow in isolation, instead hosting a constantly changing and understudied microbiome. It is known that this can have impacts ranging from nutrient uptake facilitation to competitive or pathogenic effects. The aim of our project is to investigate the effects of different carbon sources on algae cultivation and the resulting microbiome changes on microalgae growth rates, photosynthetic health, and more.

- \succ Four 1000 L outdoor raceway ponds were set up to grow Nannochloropsis sp. Co18
- \succ Two ponds were only bubbled with CO₂ (control) and two ponds (treatments) were supplied with water and a higher concentration of dissolved inorganic carbon in the form of a bicarbonate (HCO₃-)
- > Measures of algae growth (turbidity, optical density etc.), photosynthesis (chlorophyll), biomass composition (FTIR), pond conditions (temperature, pH) and more were collected over the six days growth cycle
- > Daily DNA and RNA samples were taken from each pond for downstream taxonomic and functional analysis of the hostmicrobiome interactions

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Results

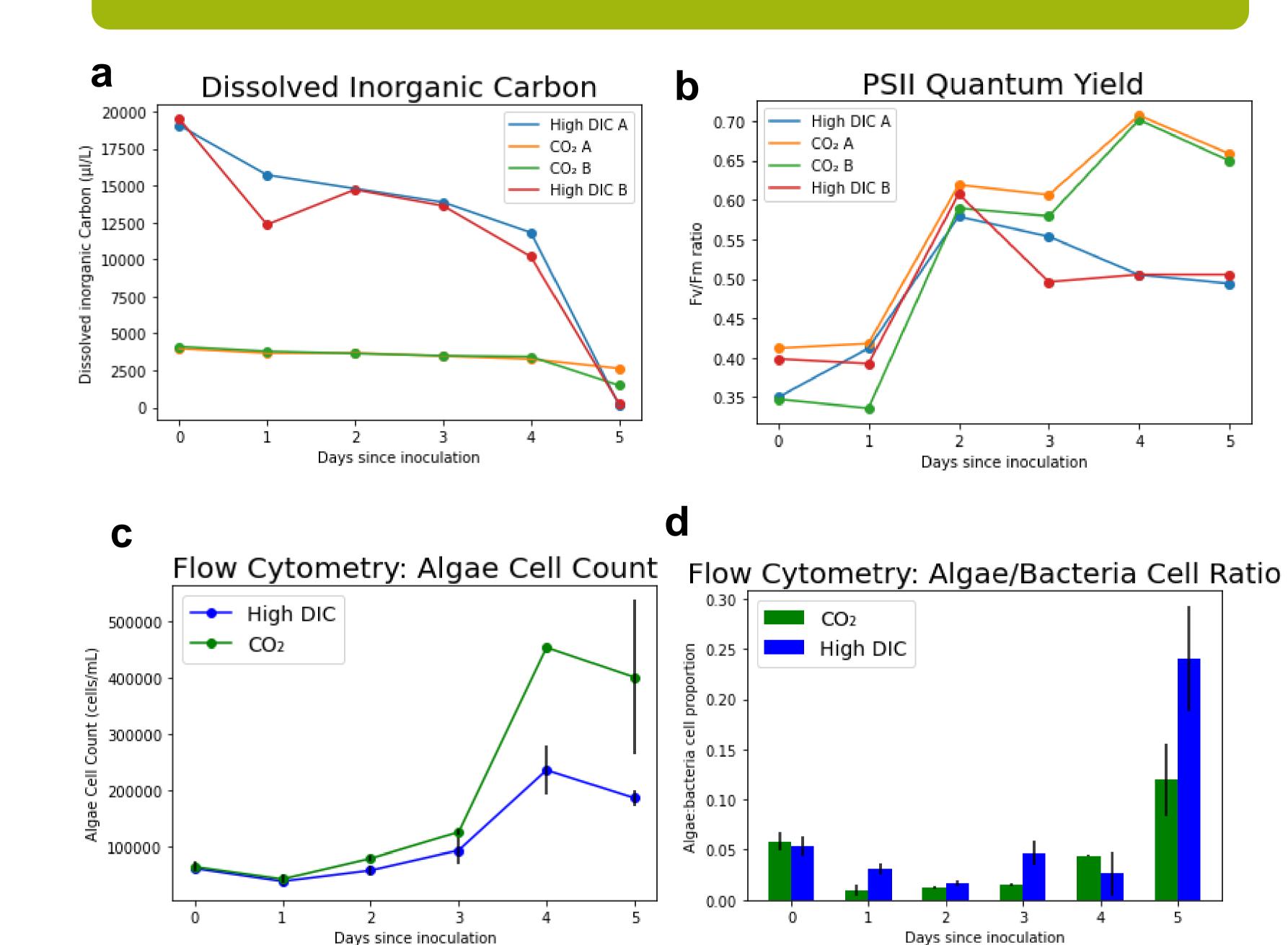
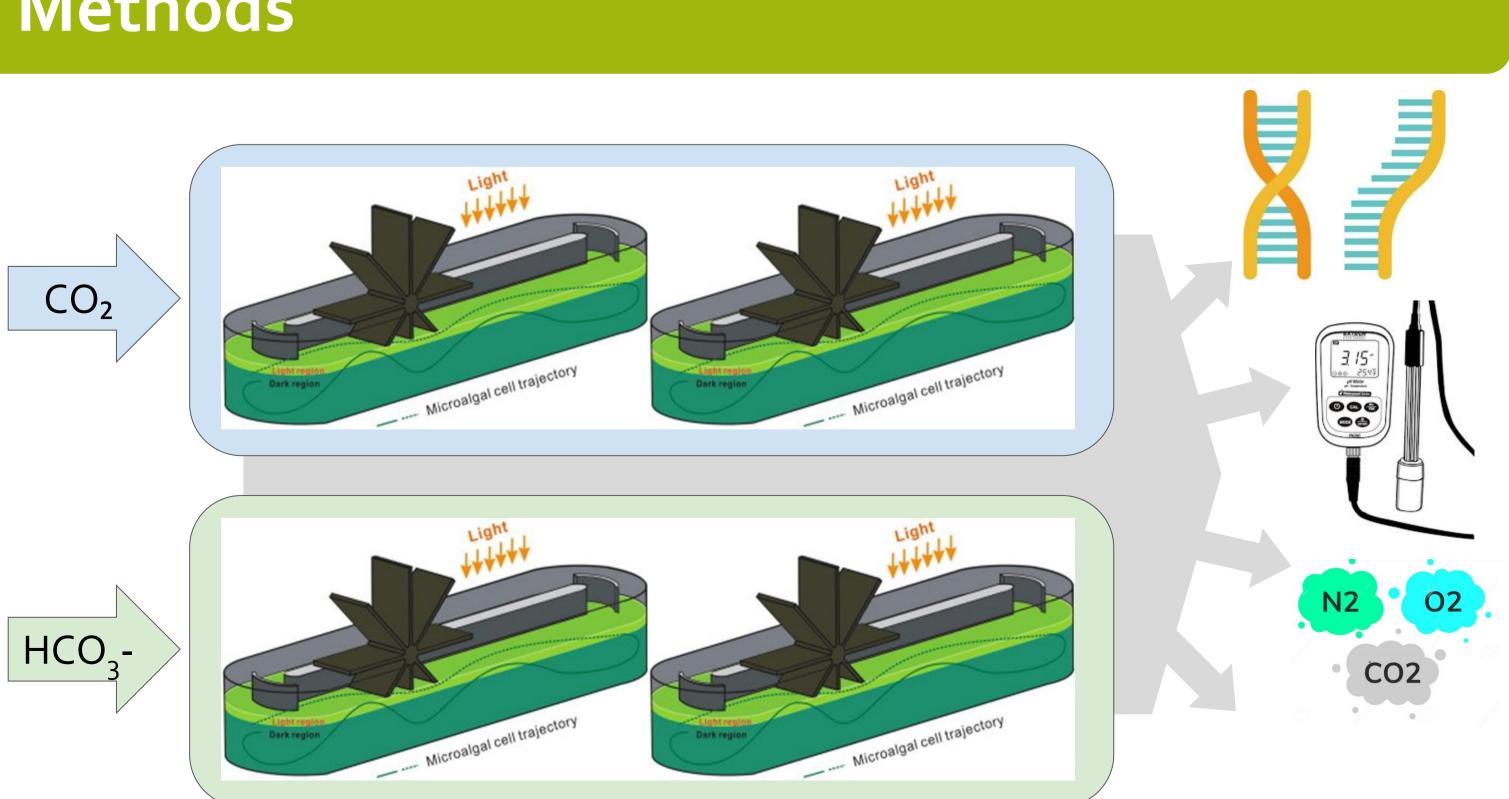
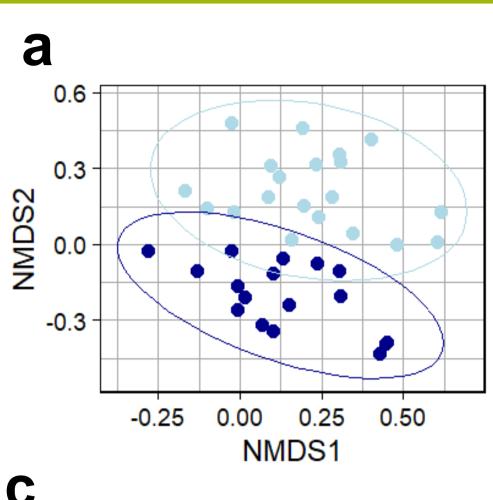


Figure 1.

- > **a**: Dissolved inorganic carbon (HCO₃ + CO₃ + CO₂) levels over time. The water with bicarbonate had higher initial DIC, but dropped to control levels by the end of the experiment > b: Photosynthetic health indicated by active fluorescence
- > c: Flow cytometry data from each pond and averaged to the plots above shows algae count grew till D4, then decreased on D5.
- > d: Algae/Bacteria ratio follows the same trend; however, bicarbonate ponds mostly had higher ratios

Methods





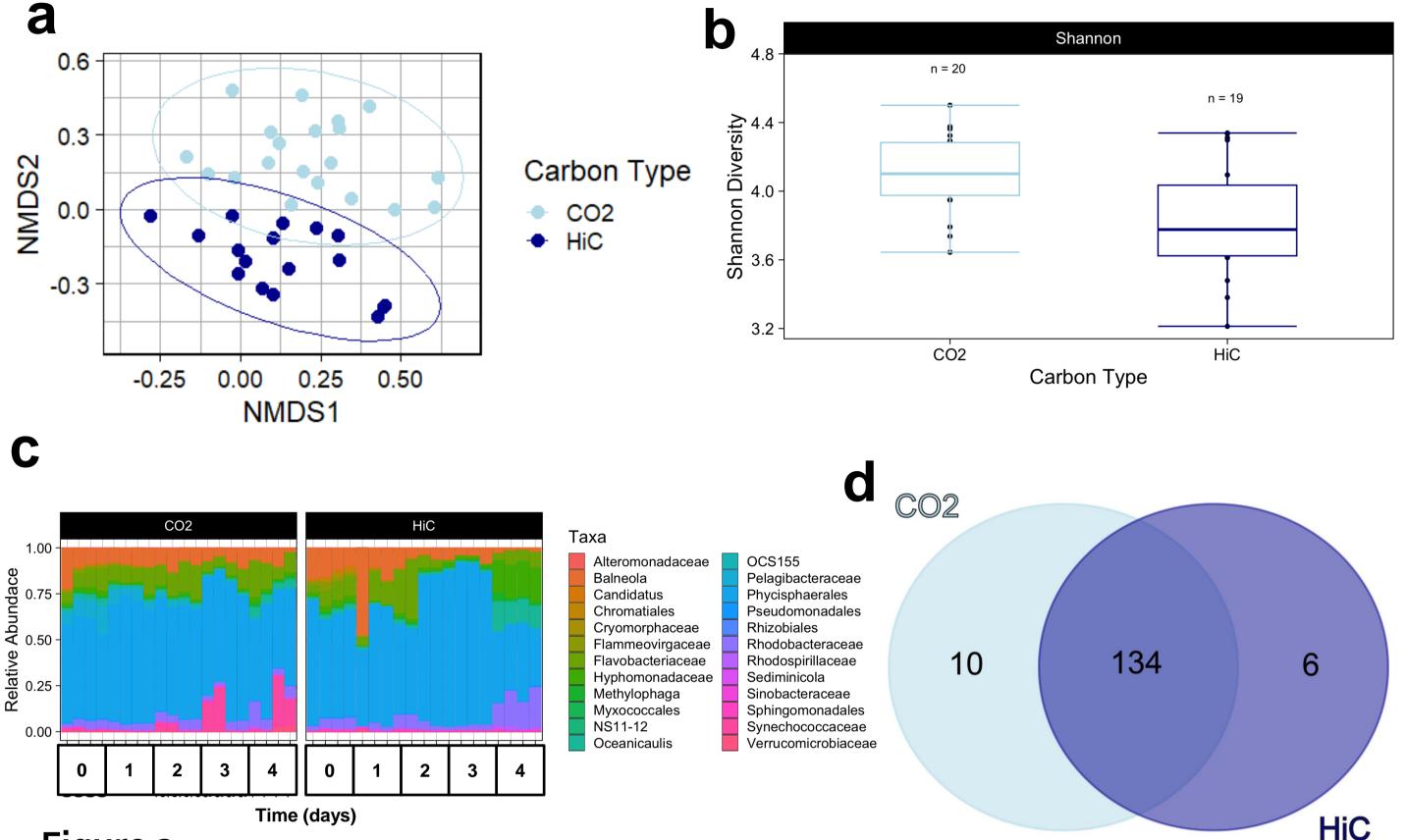


Figure 2.

- > a: NMDS ordination based on Bray Curtis dissimilarity revealed effects of carbon source on microbial community structure
- > **b**: Shannon Diversity was similar for CO₂ and HiC ponds.
- D3 onwards, which could be associated with findings in Figure 1c and 1d.
- > c: Synechococcaceae experiences a period of increase in CO2 ponds from > d:Majority of taxa were found in both conditions

Based on algae pond measurements so far:

- I. Microalgae grown with only bubbled CO₂ had higher growth rates and photosynthetic rates from day 3 onwards.
- source condition, with bacteria dropping steeply on the last day. time, and can even influence microbiome structure.
- II. Concentrations of algae and bacteria varied by time and carbon III.DIC type affects species distribution, including changes across

commercialization of sustainable algae products.

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Conclusions

- **Ongoing work/Future Diections:** continue metatranscriptomic analyses of the algae-microbiome interaction to predict functional features of each taxa of bacteria. We will also consider how this changes under pond conditions by analyzing differentially expressed genes. The work will help to inform the long term goal of optimzing algae growth by engineering microbiomes to enhance the
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