Morphological and Physiological Changes of the Marine Diatom *Thalassiosira weissflogii* Over a Growth Cycle

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INTRODUCTION

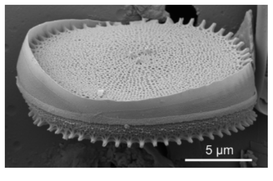
Thalassiosira weissflogii (*T. w.*) is a centric diatom, a unicellular algae species, that grows in marine environments around our globe. This organism is important for the marine food web but requires a substantial amount of nutrients to grow to high biomass. Diatoms are eukaryotic benthic or pelagic algae with silica walls as distinctive feature. These organisms vary from 3 to 500 microns in size ("Diatoms", UCL). Dense cultures can be seen to the naked eye; however, diatoms are best viewed through a stereoscopic microscope. They are autotrophic photosynthetic organisms in that they convert CO_2 into organic matter and produce O_2 from splitting the water molecule. Sunlight is required to power this cellular process. Diatoms are sensitive to environmental conditions including light, salinity, and temperature.
Indicators of a healthy diatom cell culture include:

- Increase in cell density over time (exponential increase)
- A constant cellular chlorophyll a and cell size
- A photosynthetic quantum yield between 0.6-0.8

PURPOSE

I investigated the growth, nitrate uptake, morphology and photosynthetic capacities of T. w. over a growth cycle lasting 19 days. The focus of this experiment was to observe morphological and physiological changes in this species during an artificially induced bloom in order to understand the growth behavior and carrying capacity under the given conditions. I observed three growth stages during this incubation: exponential phase, stationary phase, and death phase. The data from this experiment will be useful to be able to estimate the best timepoint to harvest cells for follow up experiments.

METHODS



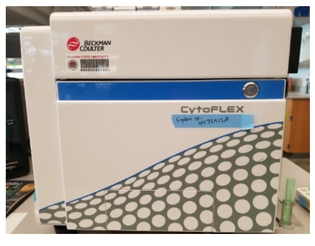


Image of *Thalassiosira weissflogii* and CytoFLEX Flow Cytometer

T.w. cultures were grown in artificial seawater containing nutrients in concentration often found in coastal systems (40 µM NO_3^-, 2.5 µM PO_4^{2-}, and 5 µM Si). Cells were grown in temperature controlled light incubators. Cell growth, cellular health and nutrient uptake were recorded using flow cytometry, variable fluorescence, and spectrometric nutrient analysis, respectively.

RESULTS

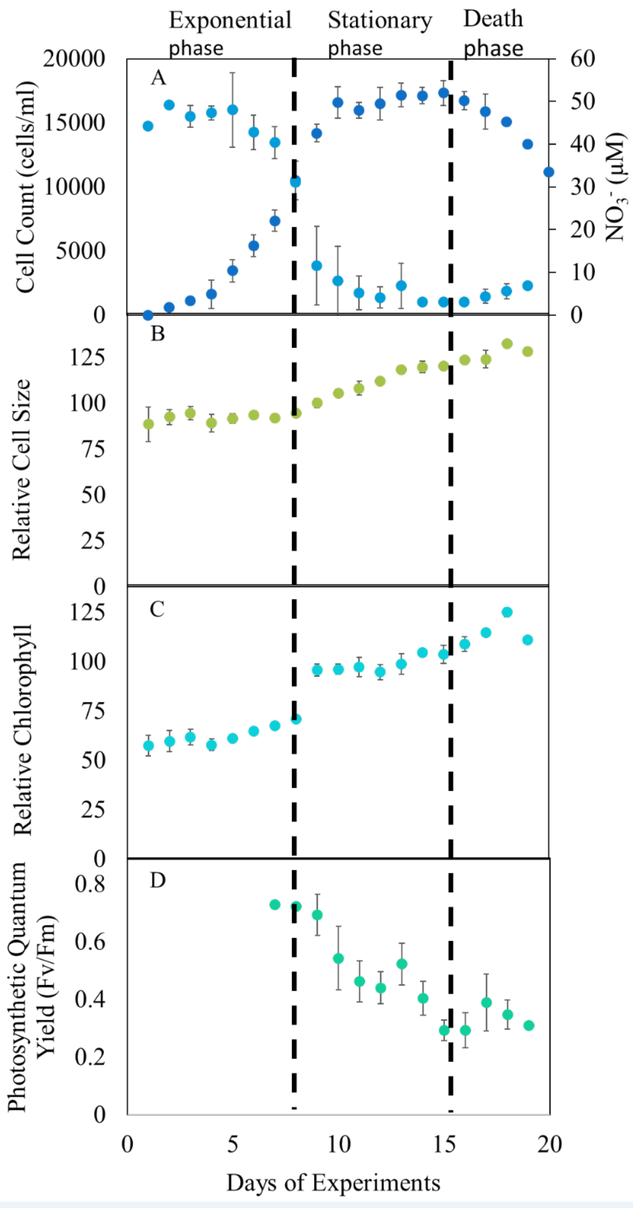


Figure 3: Growth Response Curves
A. Changes in Cell Density Over Time vs NO3 Concentrations Over Time
B. Changes in Relative Cell Size Over Time
C. Changes in Relative Chlorophyll Over Time
D. Changes in Photosynthetic Quantum Yield Over Time

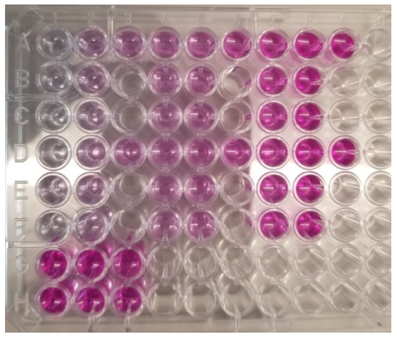
CONCLUSIONS

Specific Growth Rate µ:
$\mu = (Ln(D) - Ln(D_0)) / t$
Growth Rates Over Growth Cycle:

- Exponential Phase: specific growth rate of 0.45 d^{-1}
- Stationary Phase: specific growth rate of 0.01 d^{-1}
- Death Phase: specific growth rate of -0.1 d^{-1}

Data:

- No lag phase was observed
- Culture carrying capacity (max cell density) reached at Day 14, at 17320 cells/ml
- NO_3^- was nearly depleted by Day 11 to about 5 µM



Analysis of NO3 concentration in cultures.

I observed three growth stages during this incubation: exponential phase (Day 1-8), stationary phase (Day 10-14), and death phase (Day 15-19). Cells grew exponentially until nearly all NO_3^- was drawn down. Subsequently, the cells stopped replicating, reaching their stationary phase and changed some of their cellular properties. We found that during this phase, the diatoms grew larger (higher biovolume), increased cellular chlorophyll all while cellular health decreased. Cells did not draw NO_3^- down to completion, indicating that PO42- was likely the limiting nutrient in my incubation. During the death phase, cell density rapidly decreased, indicating a fast loss of biomass and food availability once nutrients become limiting.

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