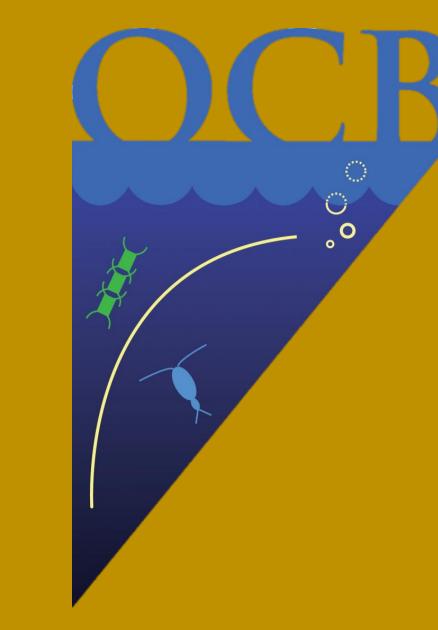
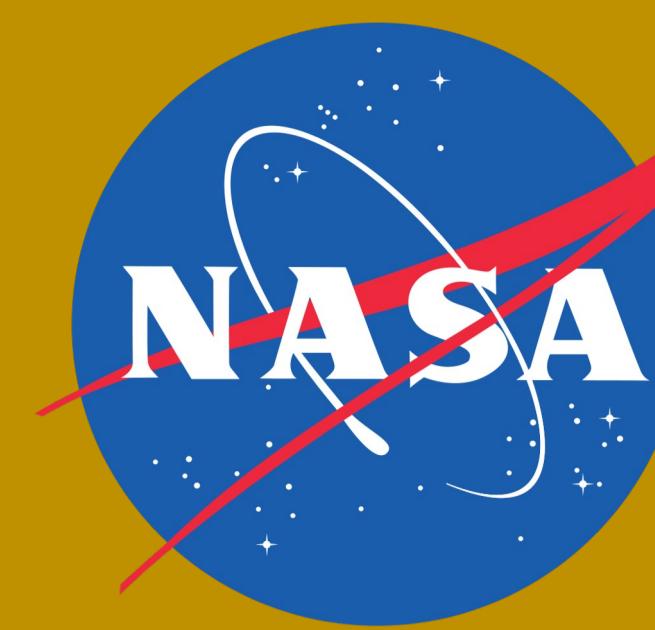
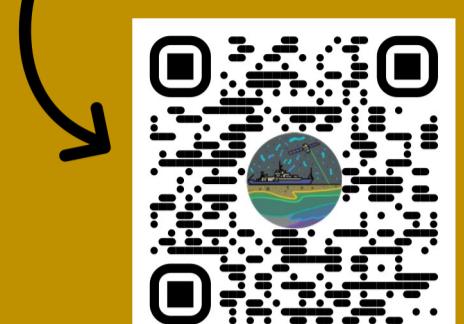


Diel b_{bp} Cycles in Cultured Phytoplankton



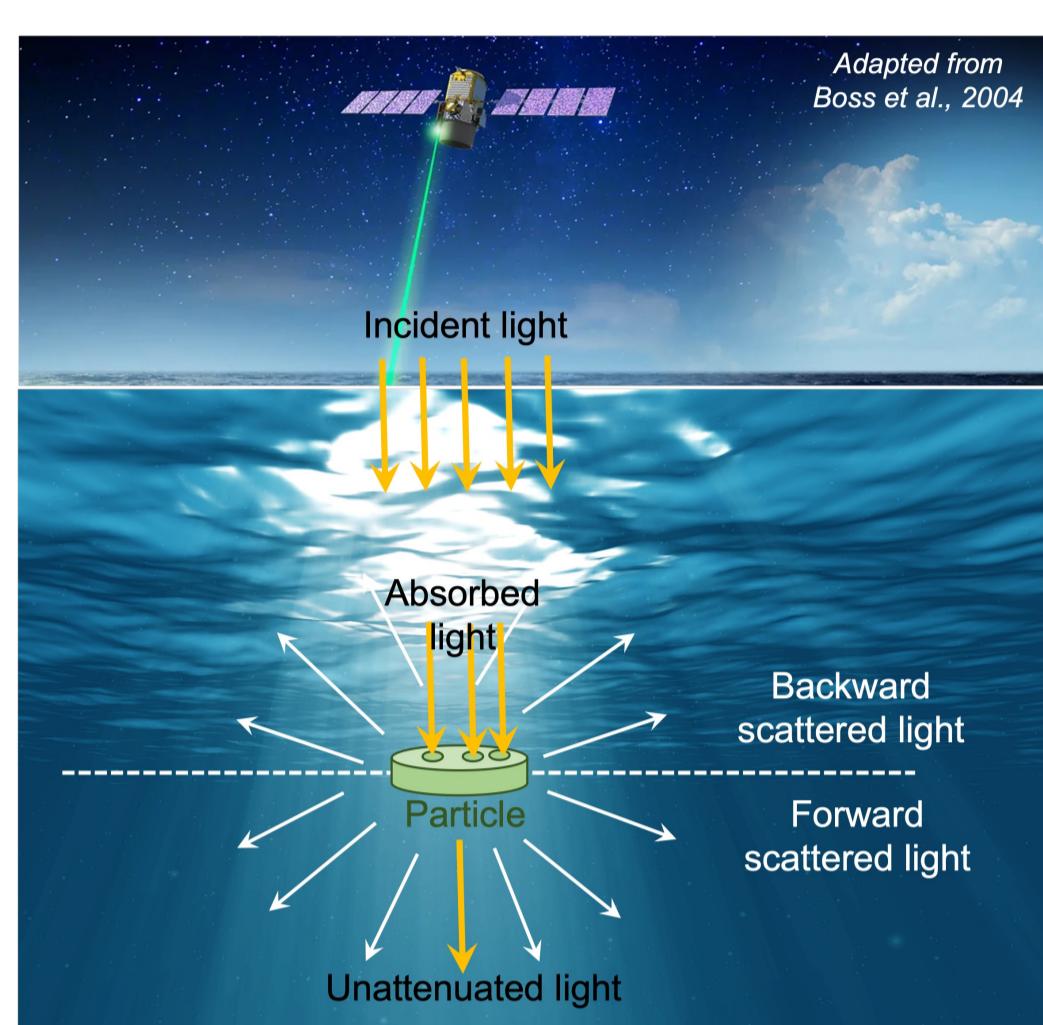
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Background & Questions

Photons scattered by particles in the backwards direction (b_{bp}) have the potential to leave the ocean surface and be detected by remote sensing. b_{bp} is a proxy for particle abundance but is also dependent on particle refractive index. Global b_{bp} has been reported to vary over diel timescales and has been shown to increase in laboratory cultures from sunrise to sunset.



- Q1: What is the contribution of phytoplankton to b_{bp} variability over a complete diel cycle?
- Q2: How does this contribution vary across species?

Figure 1. Attenuation of light in the surface ocean.

Synchronized Cell Division

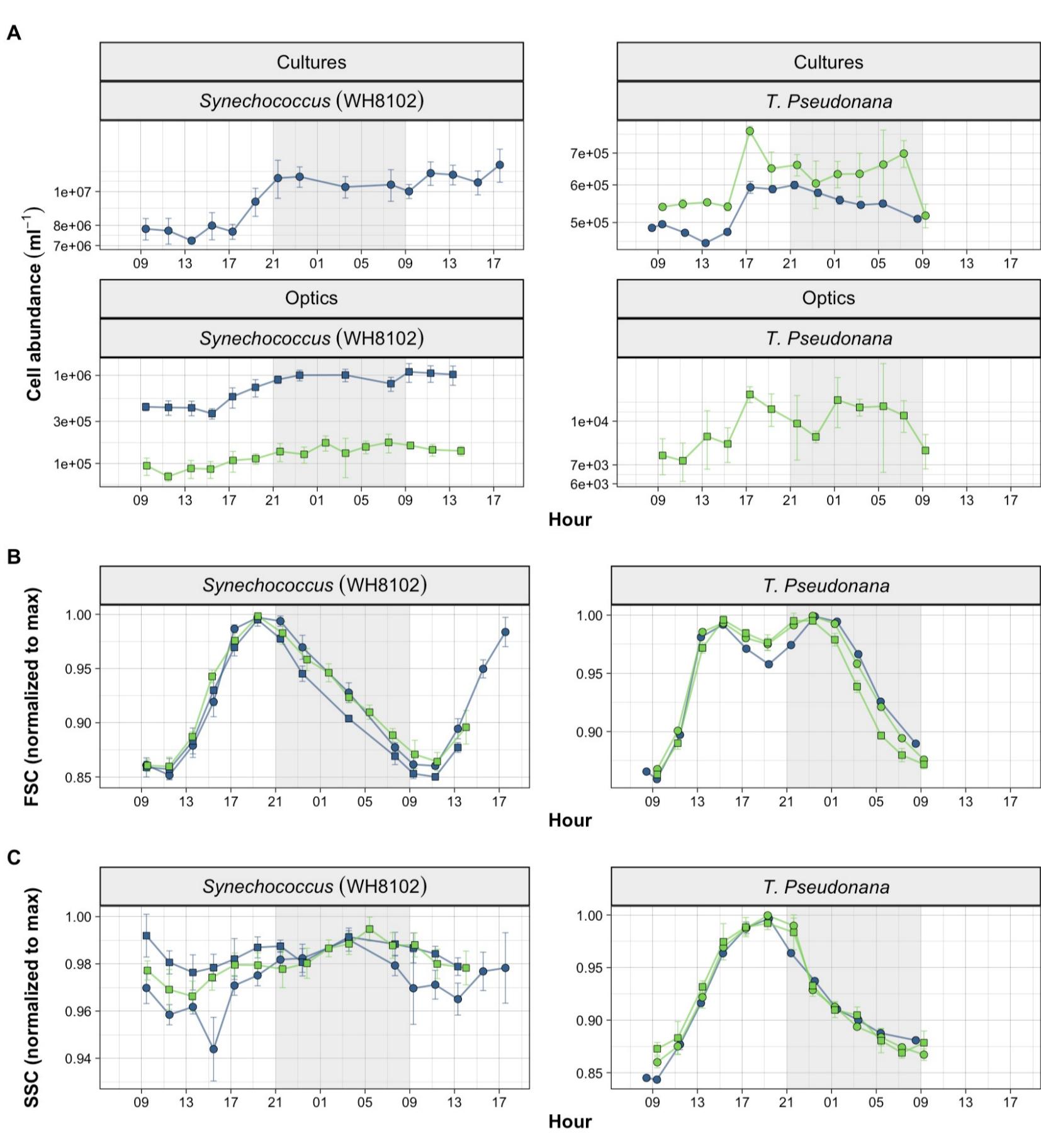


Figure 3. Cell abundance (A), forward scatter (FSC; B) and side scatter (SSC; C) per cell. Unshaded and shaded areas denote day/night (dawn = 900, noon = 1500, dusk = 2100). Error bars denote standard deviation from mean of biological replicates for each experiment.

- Both phytoplankton species displayed synchronized cell division, with SYN division ($\mu = 0.43 \text{ d}^{-1}$) occurring over shorter span than TP ($\mu = 1.1 \text{ d}^{-1}$). Note: TP cultures were diluted to starting concentrations when $\mu > 0$ (TP division between 1700 and 0900).
- FSC (cell size proxy) increased during the day and decreased during night-time division and respiration.
- SSC (proxy for cell complexity and granularity) showed similar diel cycle for TP but out of phase with division for SYN.

Photosynthetic Efficiency

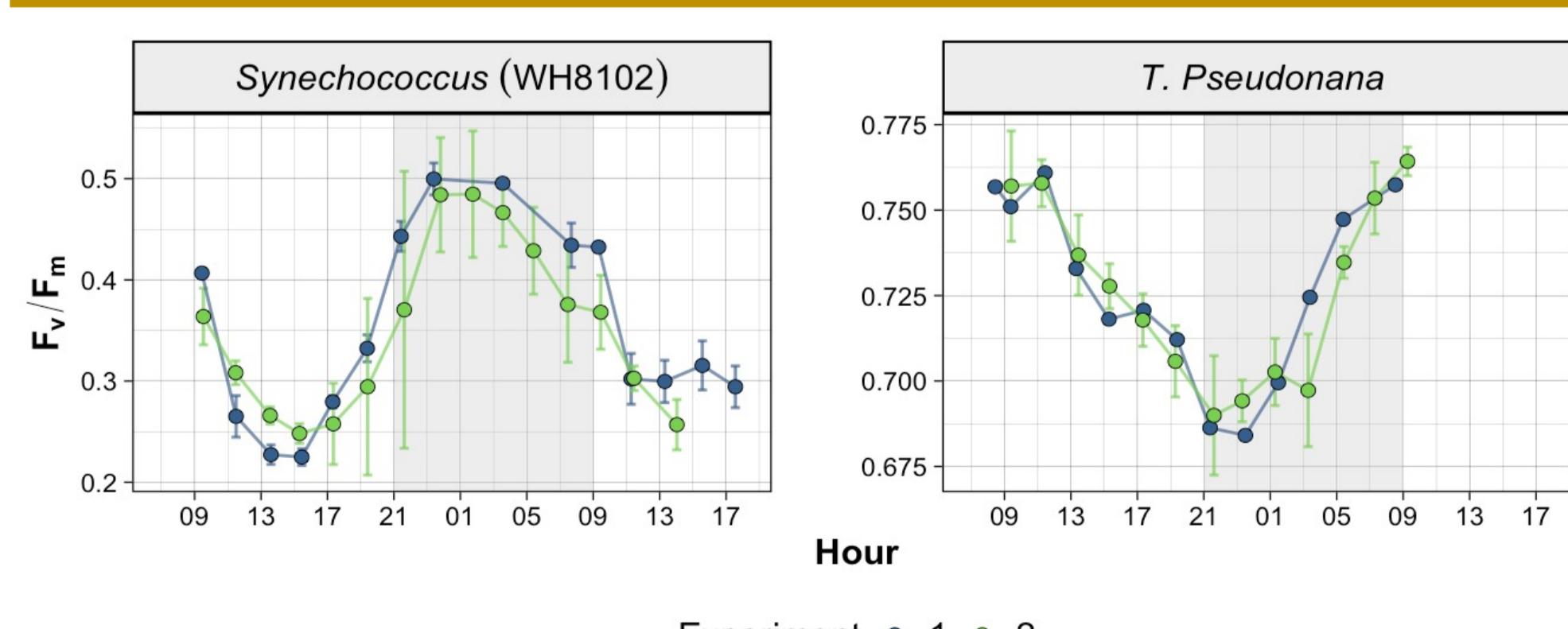


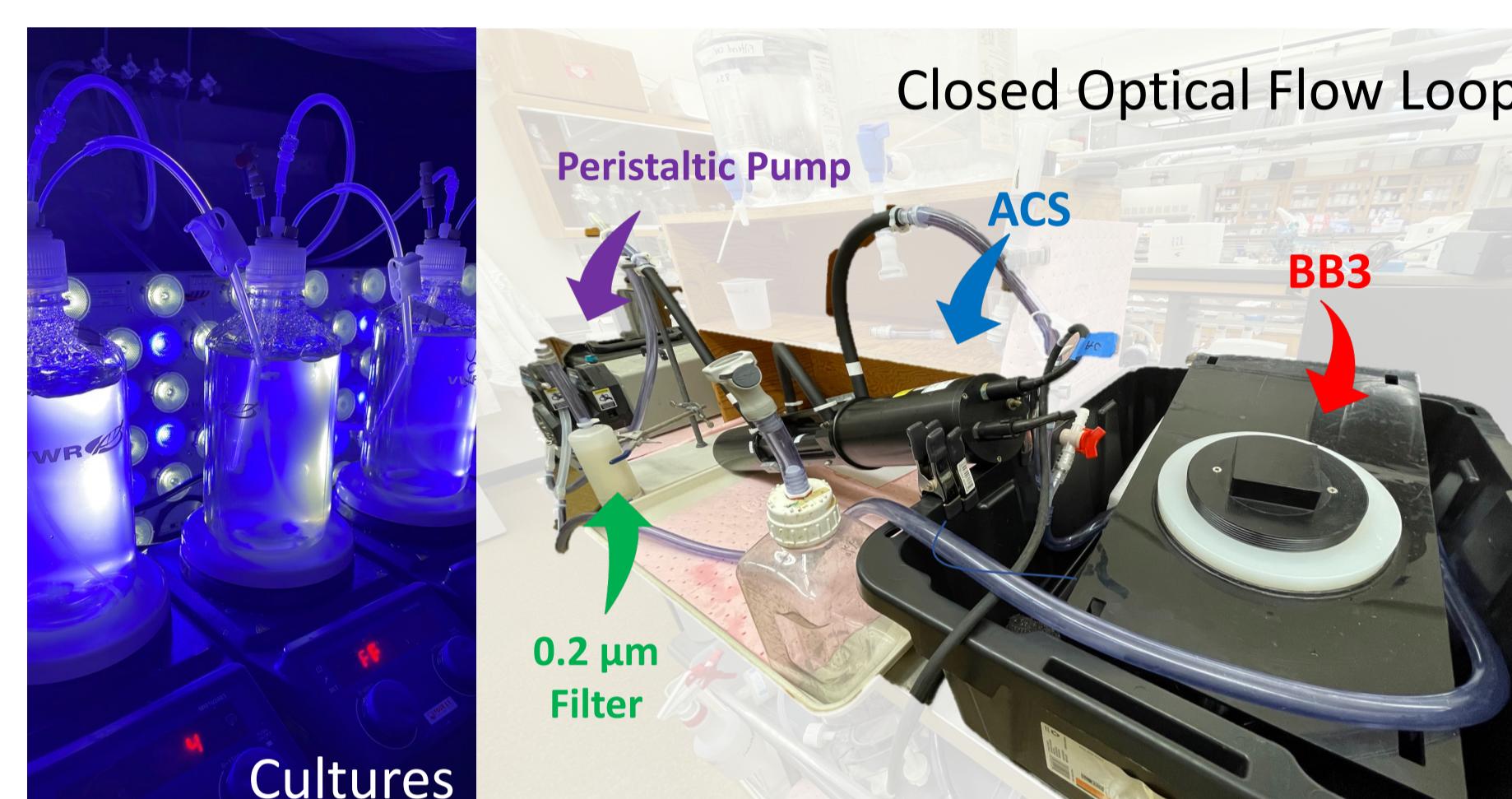
Figure 6. Variable to maximum fluorescence (F_v/F_m) of changes in response to available light for both phytoplankton. Error bars denote the standard deviation of the mean of biological replicates for each experiment.

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Hypotheses & Approach

H1: Diel variations of b_{bp} will reflect increases in phytoplankton size and organic carbon content during the day and decreases during the night due to division and respiration.

H2: The magnitude and timing of diel changes in b_{bp} will be heavily influenced by the degree of cell cycle synchronization, which can be greater for some species relative to others.



- Complete diel cycles characterized for *Thalassiosira pseudonana* (TP) and *Synechococcus* (WH8102; SYN), grown in nutrient-replete conditions at 18°C under 12:12 h sinusoidal light: dark period (maximum intensity of 400 $\mu\text{E m}^{-2} \text{s}^{-1}$).
- Particulate absorption (a_p), attenuation (c_p), and b_{bp} measurements collected using a WET Labs ACS and ECO-BB3 every 2 hours for at least 24 hours.
- Ancillary samples for cell enumeration and scattering via flow cytometry; photophysiology via fast-repetition rate fluorometry (FRRF); cell organic carbon and nitrogen; cell macromolecular composition; SEM.

Figure 2. Culture conditions (left) and sampling set up of optical instruments (right).

Diel Cycles in Optical Properties

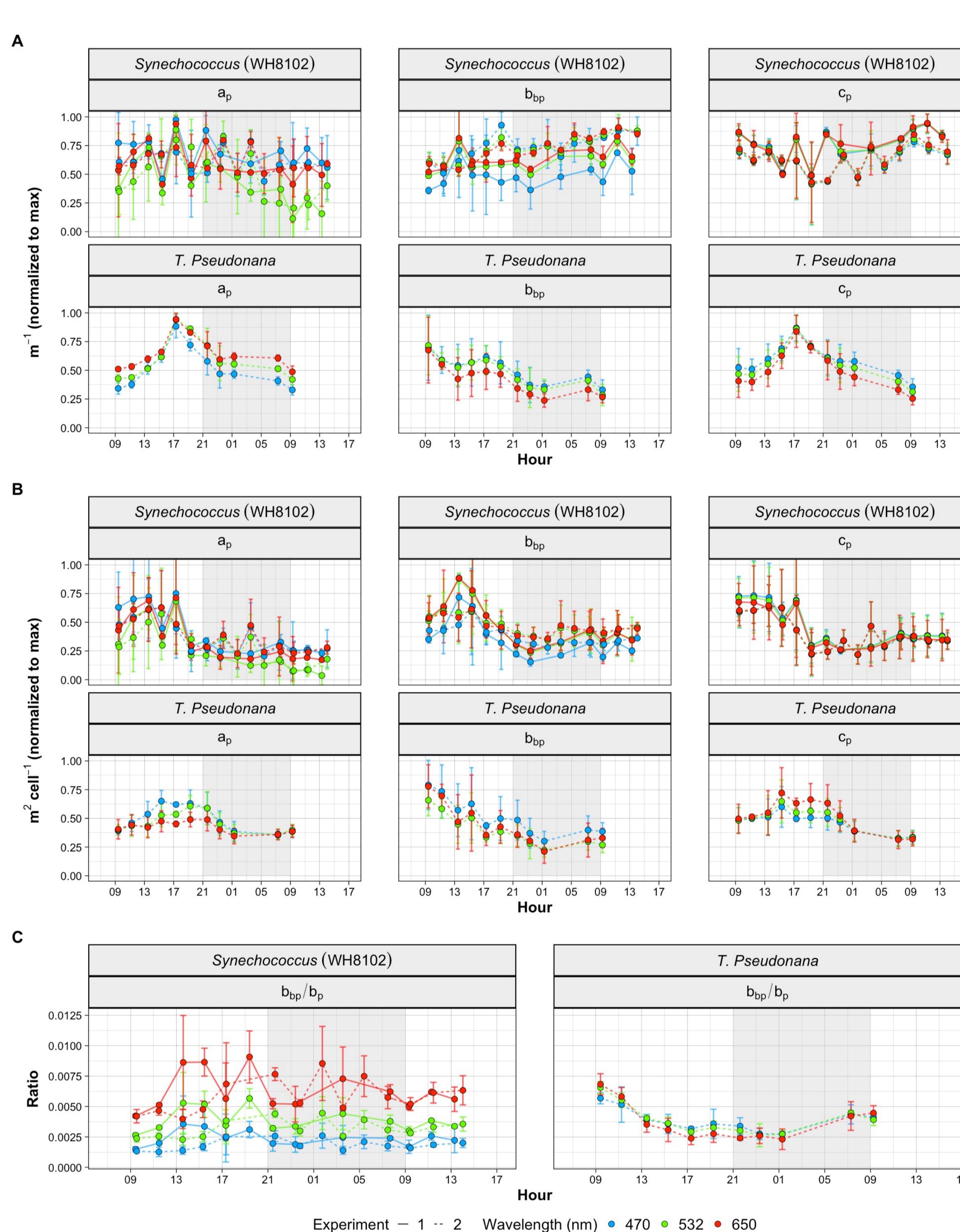


Figure 4. Bulk (A) and cell concentration-normalized (B) properties of a_p , b_{bp} , and c_p . (C) Backscattering efficiency (b_{bp}/b_p). Error bars denote the standard deviation from mean of biological replicates for each experiment.

- Bulk optical properties showed diel variability for only TP, while normalized properties show similar patterns between species.
- Coefficients are maximal prior to cell division during the day and then decrease following division, reaching minimums during the night.
- Changes in a_p and c_p are more pronounced for SYN while changes in b_{bp} are comparable between the two phytoplankton.
- b_{bp}/b_p varies over the diel cycle for TP but not for SYN.
- Little to no spectral dependence for any of the properties for either phytoplankton, except for b_{bp}/b_p for SYN.

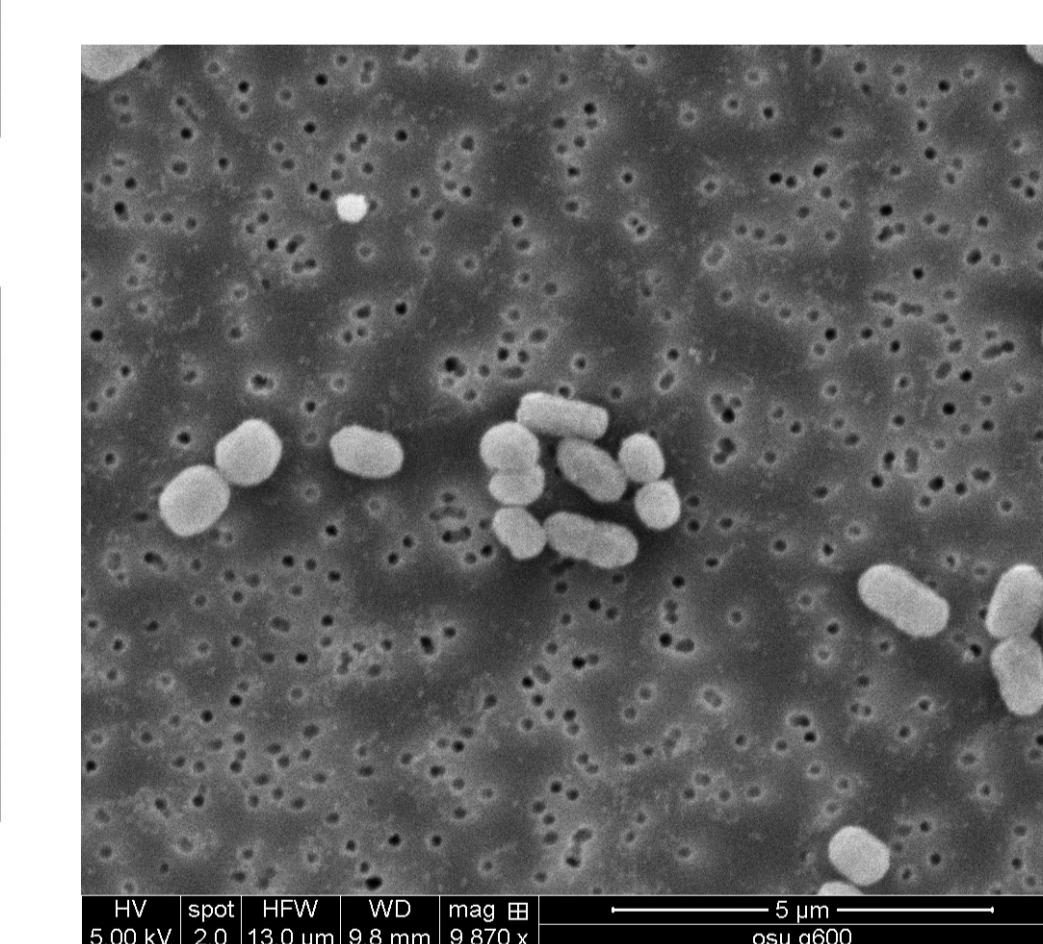


Figure 5. Scanning Electron Microscopy (SEM) image of *Synechococcus* from 0500.

a_p Spectral Decomposition & Pigment Ratios

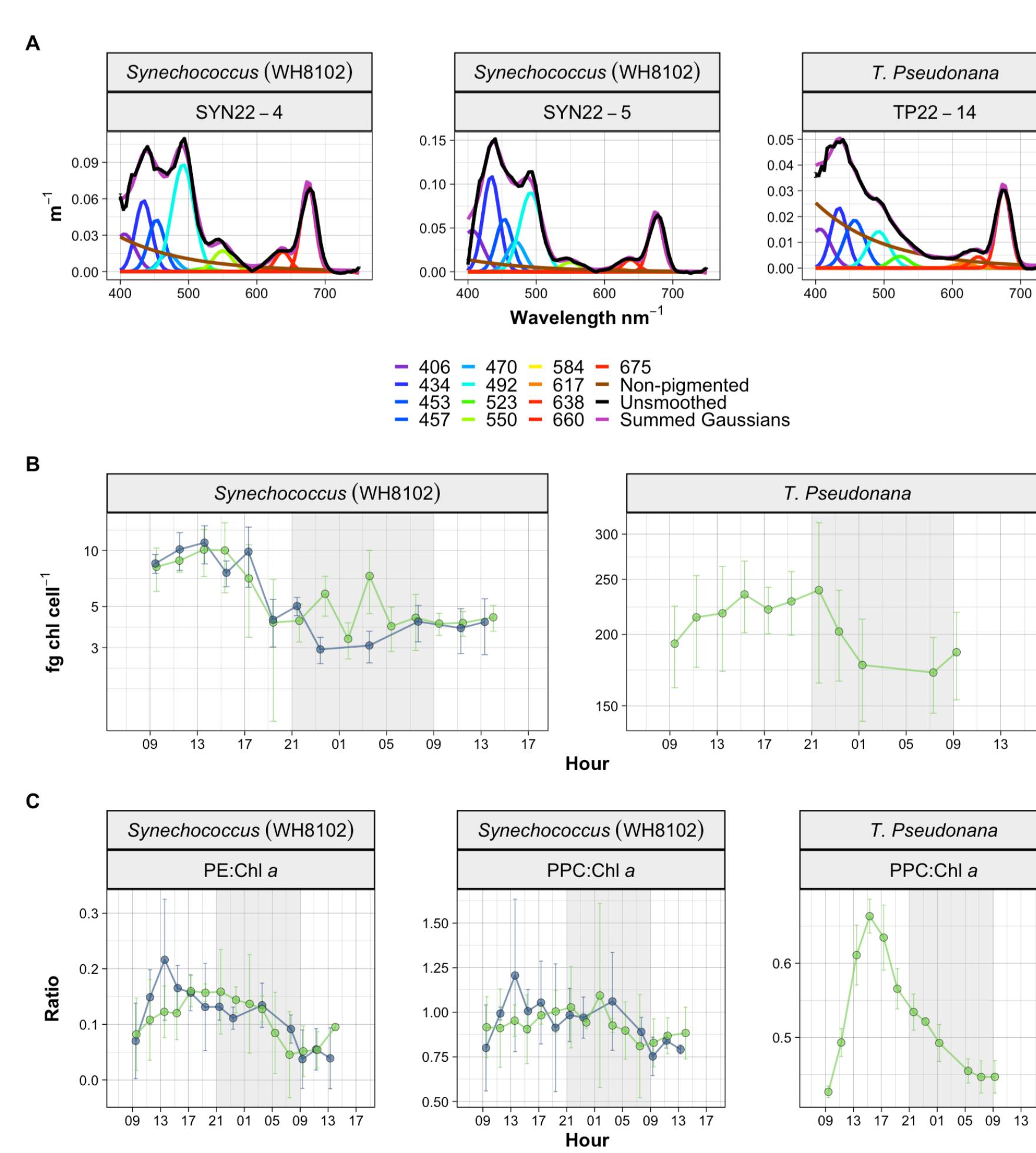


Figure 7. Examples of Gaussian decomposition of absorption spectra (A). Cell-normalized chlorophyll concentrations estimated from absorption line height at 676 nm (B). Ratios of Gaussians for phycoerythrin (PE, 550 nm), photoprotective carotenoids (PPC, 492 nm), and chlorophyll a (Chl a, 675 nm). Error bars denote the standard deviation of the mean of biological replicates for each experiment.

- Cell-normalized chlorophyll follows pattern of cell division, with a 5 h to 6 h lag for TP.
- Diel patterns in pigment ratios follow changes in light intensity and are dampened for SYN relative to TP.

Conclusions

- Diel variations in b_{bp} generally reflect changes in the cell cycle and photophysiology. However, the timing of b_{bp} peaks and valleys can be out of phase with cell abundance, photosynthetic efficiency, and pigment ratios.
- The pronounced diel changes in phytoplankton b_{bp} suggests that time of day is an important consideration for interpreting satellite b_{bp} retrievals.