June 12, 2025

Response for reviewers

We wish to thank the reviewer and Associate Editor for their continued efforts to improve this manuscript. Their comments are copied below, followed by our point-by-point responses in red.

Reviewer 1 Comments

L176-177: "Light was supplied evenly by the five actinic LEDs within the FRRF (445, 470, 505, 530, and 590 nm)…."
I suppose your FRRf LEDs were surely calibrated but all of which were at the same levels. As you showed in Appendix B, diatoms were exclusively dominant; however, there might be some optical/spectral differences between waters and between surface and subsurface waters. Although it could have minimal effects on Φe:NCP, is there any possibility that the evenly calibrated LEDs partly decouple Φe:NCP? Recent FRRf automatically tunes the excitation spectral distribution as the manufacturer says.

All lamps were calibrated against a handheld light meter prior to deployment (L177-178).

The reviewer brings up an important point about spectral corrections. We have added a new section to the discussion (copied below) highlighting the potential effects of spectral differences between instrument light sources and in-situ light environments on the relationship between ETR_{PSII}, C-fixation, and NCP.

4.3.3 Final methodological considerations: spectral corrections

As discussed above, decoupling between ETR_{PSII}, C-fixation and NCP is affected by methodological factors, including differences in the time scale of different photosynthetic processes, and the different normalizations for various measured rates (e.g. per volume, Chl or RCII). Additionally, differences in the spectral characteristics of the in-situ light environment and instrument light sources may contribute further to decoupling between ETR_{PSII}, C-fixation, and NCP.

Phytoplankton exhibit variable light use efficiencies across the photosynthetically available wavelength spectrum, due to non-uniform pigment compositions and absorption spectra across assemblages. Consequently, photosynthetic measurements are wavelength-dependent (Kyewalyanga, Platt and Sathyendranath, 1997). As a result, differences in the spectral distribution of light between the FRRF and photosynthetron incubator (used for ^{14}C uptake measurements) and the in-situ light environment could influence the stoichiometry we observed between ETR_{PSII}, C-fixation, and NCP.

In principal, spectral corrections, can be used to account for variability between instrument light sources and in-situ light environments to improve the inter-comparability of measurements (Schuback., et al., 2021; Tortell et al., 2023). These corrections require

measurements of the spectral distribution of FRRF, photosynthetron incubator, and in-situ light, and a reconstruction of photosynthesis absorption spectra based on photosynthetic pigment concentrations, determined from HPLC analysis. These corrections rely on the assumption that absorption spectra of photosynthetic pigments accurately represent the action spectra of photosynthesis, which not always the case (Kyewalyanga, Platt and Sathyendranath, 1997). In our study, we did not collect measurements of the spectral distribution of light in the euphotic zone, which would have likely varied significantly across depths and sampling sites (i.e. on-shore versus off-shore). For this reason, we are not able to spectrally correct in vitro measurements of ETR_{PSII} and ^{14}C-uptake to in-situ spectral fields for more direct comparison with NCP. As a result, spectral differences between instrument light sources and the in-situ light environment could contribute to some of the observed variability between ETR_{PSII}, C-fixation and NCP in this study. As for the comparison of FRRF and ^{14}C data, spectral corrections would affect the absolute magnitude of $\phi_{e:C}/n_{PSII}$, but these corrections would yield a station-specific scalar and would not affect the hyperbolic relationship observed between ETR_{PSII} and C-fixation.

The best approach to minimize the influence of spectral variability is to match the spectral properties of instrument light sources to ambient light fields. However, this remains challenging for high-resolution underway applications across varying spectral environments.

Also is your Φe:NCP prediction successful due to the diatom-dominated community? Or would be successful even in various algal communities? Any possible ideas?

We hypothesized that the strength of the correlation between ETR-derived GP and NCP was primarily due to similar dependencies on [Chl]. See L892 – 925 copied below.

'Despite the inherent dependency of net oxygen production on gross oxygen production, the strength of the correlation between NCP and ETR_{PSII} and the consistency of ETR_{PSII}:NCP across subregions is surprising given the multitude of methodological and phsyiological factors that can uncouple these rates (Fig 7). However, the derivations of NCP and GP both have similar dependencies on mixed layer Chl concentration. To obtain FRRF-derived GP estimates in comparable units of mmol O_2 m^{-2} d^{-1}, we multiplied in-situ ETR_{PSII} by mixed layer Chl concentrations (Eq 5). While mixed layer Chl concentrations are not explicitly included in NCP calculations (Sect 2.6), biomass is expected to be a primary driver of bulk productivity. If Chl-normalized NCP is instead compared against GP expressed in units of mmol O_2 Chl^{-1} d^{-1}, the correlation between 24h binned and instantaneous NCP and ETR_{PSII} estimates decrease to ρ = 0.22 and 0.35, respectively. We therefore conclude that it remains challenging to derive gross and net carbon fluxes from FRRF measurements alone, but paired ETR_{PSII} and Chl measurements can provide useful constraints for NCP estimates.'

Thank you for providing your transcriptomic data. It looks interesting %diatom was high with high %protists and vice versa. However, I'm curious what the-axis of Fea1 bars indicates. The "counts" indicated "calibrated" counts referenced reads of "diatoms" like

“TPM reference to the total diatom reads”? The Y-axis needs more explanation/clarification,

We have added a few details to the figure caption. Counts were normalized using DESeq2’s median of ratios method (Love, Huber and Anders, 2014), and refer to reads mapping to diatoms.

Editor Comments

With the next revision, please re-name the appendix figures to B1 and C1.

Figure labels updated.