### 2008 Circumpolar Flaw Lead (CFL) System Study

Kevin Arrigo Department of Environmental Earth System Science 473 Via Ortega Stanford University Stanford, CA 94305-4216 Phone (650) 723-3599 Fax (650) 498-5099 arrigo@stanford.edu

### **Photosynthesis-Irradiance Incubations**

Photosynthetic characteristics were obtained from photosynthesis-irradiance (P-E) measurements using a modification of the <sup>14</sup>C bicarbonate technique of Lewis and Smith (1983) as modified by Arrigo et al. (2010). P-E incubations were carried out at 2°C and at 20 irradiances ranging from 0 to 635 µmol quanta m<sup>-2</sup> s<sup>-1</sup>. The incubator was an aluminum block containing illumination chambers to hold the incubation vials and channels through which coolant was continuously circulated. Even illuminator (Lumenyte International Corporation, model DMX512) fitted with a 150 W tungsten-halogen lamp. Total photosynthetically available radiation (PAR, 400-700 nm) within each illumination chamber was measured using a Spherical Micro Quantum Sensor US-SQS (Heinz Walz, GmBH). Spectral irradiance,  $E(\lambda)$ , was measured from 300-800 nm using a spectroradiometer (Analytical Spectral Devices, FieldSpec).

For each P-E curve, a 50 ml sample of stock culture was placed in a pre-chilled, 200 ml plastic beaker and spiked with 0.925 MBq  $H^{14}CO_3$  to obtain a final activity of 0.019 MBq ml<sup>-1</sup>. After thorough but gentle mixing, the spiked sample was distributed in 2 ml aliquots to 23 pre-chilled 20 ml plastic (polyethylene terephthalate) scintillation vials. Twenty of these vials were placed in individually illuminated chambers within the incubator and the incubation was initiated by turning on the light source. The contents of the three remaining vials (time-zero samples) were acidified with 100 µl of 6N HCl to drive off inorganic carbon and determine the background particulate radioactivity levels. To determine total radioactivity in the spiked sample, three 100 µl aliquots of spiked sample were added to scintillation vials containing 0.5 ml filtered seawater, 5 ml of scintillation fluid (EcoLume) and 100 µl of ethylamine and then assayed for radioactivity by liquid scintillation counting (Perkin Elmer WinSpectral 1414). Incubations were terminated after 1 h by turning off the light source and adding 100 µl of 6N HCl to each vial. All acidified samples were gently shaken for a minimum of 12 h to drive off radioactive inorganic carbon and then neutralized with 100 ml of 6N NaOH and mixed with 10 ml scintillation fluid. Radioactivity was determined by liquid scintillation counting.

The P-E parameters were derived from a fit of P-E data to the equation of Platt et al. (1980)

$$P^* = P_s^* (1 - e^{-\alpha^* E/P_s^*}) e^{-\beta^* E/P_s^*} - P_0^*$$
(1)

where  $P^*$  is the photosynthetic rate at irradiance E,  $P_*$  is the light-saturated photosynthetic rate in the absence of photoinhibition,  $\alpha^*$  is the initial slope of the P-E curve,  $\beta^*$  is a measure of photoinhibition, and  $P_0^*$  is the CO<sub>2</sub> uptake or release at E = 0 µmol quanta m<sup>-2</sup> s<sup>-1</sup>. The superscript \* indicates that the term is normalized to Chl *a*. For POC-normalized parameters, the superscript \* is replaced with *C*.

 $P^*$ , the maximum realized photosynthetic rate (photosynthetic capacity), was calculated as

$$P_m^* = P_s^* \left( \frac{\alpha^*}{\alpha^* + \beta^*} \right) \left( \frac{\beta^*}{\alpha^* + \beta^*} \right)^{\beta^*/\alpha^*}.$$
(2)

The photoacclimation parameter  $E_k$ , is calculated as  $P_{\alpha}^*/\alpha^*$ .

#### **Particulate absorption**

Specific absorption coefficients. Microalgae were filtered under low vacuum pressure onto glass fiber filters (Whatman GF/F) and particulate absorption spectra  $(a_p(\lambda), m^{-1})$  were measured between wavelengths of 300 nm and 800 nm using a Perkin-Elmer Lambda 35 spectrophotometer following the method of Mitchell and Kiefer (1988). Detrital absorption spectra  $(a_d(\lambda), m^{-1})$  were determined for each  $a_p(\lambda)$  spectra using the methanol extraction technique of Kishino et al. (1985). All  $a_p(\lambda)$  and  $a_d(\lambda)$  spectra were corrected for optical path amplification using the procedure of Mitchell and Kiefer (1988) and the coefficients of Bricaud and Stramski (1995). Chl *a*-specific phytoplankton absorption spectra  $(a^*_{ph}(\lambda), m^2 mg^{-1} \text{ Chl } a)$  were calculated by subtraction of  $a_d(\lambda)$  from  $a_p(\lambda)$  (Roesler et al. 1989) and normalized to fluorometrically determined Chl *a* concentrations.

The spectrally weighted mean Chl *a*-specific absorption coefficient for phytoplankton ( $\overline{a}^*$ , m<sup>2</sup> mg<sup>-1</sup> Chl *a*) was calculated using the equation

$$\overline{a}^{*} = \frac{\sum_{400}^{700} a_{\text{ph}}^{*}(\lambda) E(\lambda)}{\sum_{400}^{700} E(\lambda)}$$
(3)

where  $E(\lambda)$  (µmol quanta m<sup>-2</sup> s<sup>-1</sup>) is the spectral irradiance of the P-E incubator.

Determination of the maximum quantum yield of photosynthesis ( $\Phi_m$ ) depended upon the light level at which the maximum  $\Phi_m$  was measured. Most models assume that  $\Phi_m$  is achieved at the lowest irradiances, although this assumption does not always hold (Johnson and Barber 2003). Therefore, we first determined  $\Phi$  at each photosynthetron light level ( $\Phi_E$ ) from  $\vec{a}^*$  and the Chl *a*-normalized CO<sub>2</sub> uptake ( $P^*$ ) using the equation presented in Johnson and Barber (2003)

$$\Phi_E = \frac{P_E^*}{43.2 \ \bar{a}^* E} \tag{4}$$

where  $\Phi_E$  is the calculated quantum yield of photosynthesis at each of the 20 irradiance levels (*E*) produced in the photosynthetron,  $P^*_E$  is the photosynthetic rate at irradiance *E*, and 43.2 is a unit conversion factor. When the assumption that  $\Phi_m$  was maximal at the lowest light level was not met,  $\Phi_m$  was chosen as the maximum value for  $\Phi_E$  attained within the range of values of *E* used in the photosynthetron. When  $\Phi_E$  was inversely related to irradiance, we calculated  $\Phi_m$  using the equation

$$\Phi_m = \frac{\alpha^*}{43.2 \ \overline{a^*}}.$$
(5)

Unit Definitions:  $P_{m}^{*}: mg C mg^{-1} Chl a h^{-1}$ Alpha\* ( $\alpha$ \*) and Beta\* ( $\beta$ \*): mg C mg<sup>-1</sup> Chl a h<sup>-1</sup> (umol quanta m<sup>-2</sup> s<sup>-1</sup>)<sup>-1</sup> Ek: umol quanta m<sup>-2</sup> s<sup>-1</sup> Quantum Yield ( $\Phi_E$  or  $\Phi_m$ ): mol C mol<sup>-1</sup> quanta Mean specific absorption coefficient ( $\bar{a}$ \*): m<sup>2</sup> mg<sup>-1</sup> Chl a

## **Sample Type Definitions:**

UISW: Under ice surface water (<2 m deep) UI SCM: Under ice subsurface chlorophyll maximum OW SCM: Open water subsurface chlorophyll maximum

# References

- Arrigo, KR, Mills MM, Kropuenske LR, van Dijken GL, Alderkamp AC, Robinson DH (2010) Photophysiology in two major Southern Ocean phytoplankton taxa: Photosynthesis and growth of *Phaeocystis antarctica* and *Fragilariopsis cylindrus* under different irradiance levels. Integr. Comp. Biol., 50(6): 950-966, doi:10.1093/icb/icq021.
- Bricaud, A, Stramski D (1995) Spectral absorption coefficients of living phytoplankton and nonalgal biogenous matter: A comparison between the Peru upwelling area and the Sargasso Sea. Limnol Oceanogr 35(2):69-75
- Johnson Z, Barber RT (2003) The low-light reduction in the quantum yield of photosynthesis: potential errors and biases when calculating the maximum quantum yield. Photosynthesis Res 75:85-95
- Kishino M, Takahashi N, Okami N, and Ichimura S (1985) Estimation of the spectral absorption coefficients of phytoplankton in the sea. Bull Mar Sci 37(2):634-642
- Lewis MR, Smith JC (1983) A small volume, short-incubation-time method for measurement of photosynthesis as a function of incident irradiance. Mar Ecol Prog Ser 13:99-102
- Mitchell BG, Kiefer DA (1988) Chlorophyll *a* specific absorption and fluorescence excitation spectra for light-limited phytoplankton. Deep Sea Res 35:639:689
- Platt T, Gallegos CL, Harrison WG (1980) Photoinhibition of photosynthesis in natural assemblages of marine phytoplankton. J Mar Res 38:687-701
- Roesler CS, Perry MJ, Carder KL (1989) Modeling in situ phytoplankton absorption from total absorption-spectra in productive inland marine waters. Limnol Oceanogr 34(8):1510-1523