

# Fasttracka II

## Advanced Fast Repetition Rate Fluorometer



CTG's Fasttracka II sensor is a high performance, Fast Repetition Rate Fluorometer (FRRF), which is suitable for use in a wide range of applications; from the assessment of primary production within the worlds oceans to environmental monitoring within reservoirs and rivers.

The Fasttracka II sensor incorporates significant improvements over the already successful Mk I unit in every area. Performance enhancements include a much higher photon irradiance (PI), more even illumination of the sample, substantially improved signal collection optics and greatly enhanced programmability.

### APPLICATIONS

- Monitoring photosynthetic performance in phytoplankton (oceanography, limnology)
- Primary production studies (oceanography)
- Toxicity monitoring
- Water quality monitoring (reservoirs, rivers and streams)
- Bloom detection
- Environmental monitoring of phytoplankton populations (water supplies, fish farming)



Fasttracka II with profiling cage and dark chamber



Fasttracka II sensor being deployed in a reservoir

### FEATURES

- Advanced programmability, with the facility to concatenate up to 10 protocols with up to 5 blocks of flashlets in each protocol
- Straightforward implementation of both Single Turnover (ST) and Multiple Turnover (MT) modes
- Suitable for profiling and in towed vehicle systems, where the ST mode is essential
- Facility to incrementally increase the interval between flashlets, between 0 and 100%, for improved relaxation kinetics in both ST and MT modes
- Normalisation of data throughout the full dynamic range (data acquired at different gain levels) is possible, thanks to a newly developed calibration method
- Options for both PAR and depth sensors
- Monitors both LED output and photomultiplier sensitivity at each flashlet, thereby minimising measurement artefacts
- Near surface operation possible, even under high ambient light conditions
- High rejection of ambient light
- Accepts supply voltage of between 9 and 72 V
- Large dynamic range (0 to 600  $\mu\text{g l}^{-1}$ )
- Fast data download (up to 900 kb) to an RS232 or USB serial port

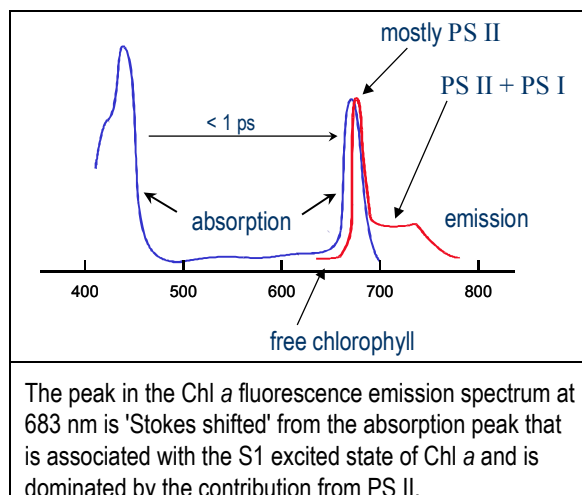
## BACKGROUND INFORMATION

### General principles

Active chlorophyll *a* (Chl *a*) fluorometry provides a non-destructive and minimally intrusive method for probing oxygenic photosynthesis, in general, and the functioning of photosystem II (PS II), in particular.

Within PS II, Chl *a* fluorescence competes directly with photochemistry and non-radiative decay for excitation energy. By measuring Chl *a* fluorescence, it is possible to gain information about the other two processes.

Chl *a* is present within both PS II and PS I. The Fast<sup>tracka</sup> II defines the Chl *a* fluorescence signal using a narrow bandpass filter, centred at 685 nm, which minimises contamination of the PS II signal from PS I and free Chl (see figure).



### The Multiple Turnover (MT) method

Fluorometers that employ the multiple-turnover method use a relatively long saturating pulse of high photon irradiance (PI) to drive the yield of photochemistry close to zero, through multiple turnovers of PS II: typically, a few hundred ms at several thousand  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ . By comparing the fluorescence signal before the pulse is applied ( $F_o$  or  $F'$  with dark or light-adapted material, respectively) with the highest fluorescence signal achieved during the saturating pulse ( $F_m$  or  $F_m'$  with dark or light-adapted material, respectively) it is possible to estimate the yield of PS II photochemistry, as  $1 - F_o/F_m$  or  $1 - F'/F_m'$  (often written as  $F_v/F_m$  or  $F_q'/F_m'$ , where  $F_v$  and  $F_q'$  are  $F_m - F_o$  and  $F_m' - F'$ , respectively).

The multiple-turnover method has a number of weaknesses:

- A multiple-turnover measurement typically takes several hundred ms. When profiling or attached to a towed vehicle, this is very likely to mean that the multiple-turnover pulse is spread over a long trail of sample, such that  $F_m$  or  $F_m'$  is never reached. This could result in a very significant underestimate of PS II photochemical efficiency
- Photosynthesis takes tens of seconds to 'recover' from the application of a multiple-turnover pulse. This limits the frequency at which measurements can be taken from a single sample
- No information about the absorption cross-section of PS II can be derived from multiple-turnover data

### The Single Turnover (ST) method

Fluorometers that employ the single-turnover method use a very short saturating pulse of very high PI to drive the yield of photochemistry close to zero, through near-simultaneous turnover of all PS II centres: typically, 100  $\mu\text{s}$  at 20 000  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ . The yield of PS II photochemistry can then be calculated, in the same way as with the multiple-turnover method.

### The Fast Repetition Rate (FRR) technique

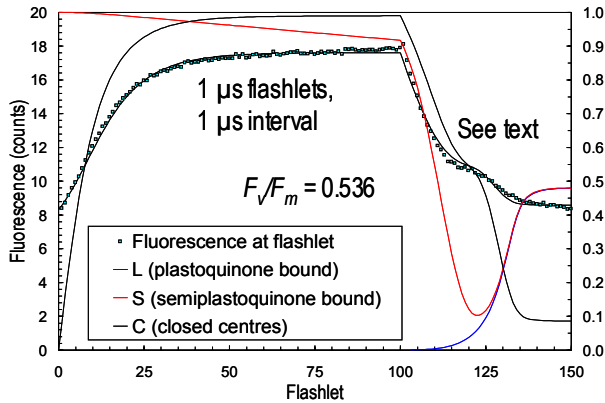
The implementation of the FRR technique within the Fast<sup>tracka</sup> II sensor maximises both the accuracy and the amount of information that can be derived from the Chl *a* fluorescence signal, by incorporating the following design features:

- An array of short (1  $\mu\text{s}$ ) measuring 'flashlets' are used both to excite Chl *a* fluorescence and provide the actinic illumination required to achieve  $F_m$  or  $F_m'$ , by driving the yield of PS II photochemistry close to zero
- The fluorescence signal is integrated over the entire duration of each flashlet
- The output from the LEDs is also integrated over the entire duration of each flashlet
- Any drift in the sensitivity of the photomultiplier tube used to record the Chl *a* fluorescence signal, or the photodiode used to monitor the output from the LEDs, is tracked by sampling between flashlets.

Overall, this approach maximises the signal to noise ratio, whilst minimising measurement artefacts.

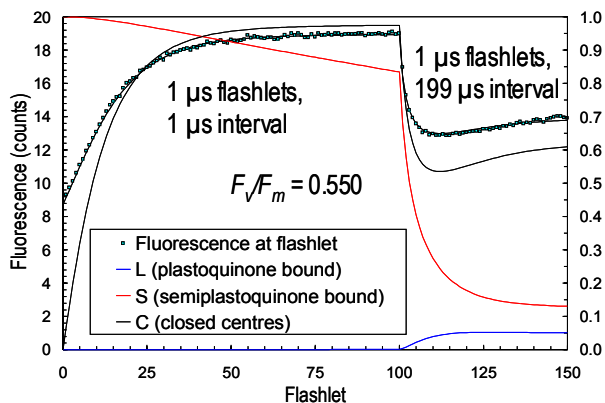
Typically, an FRR ST saturation phase is formed of 100 x 1  $\mu\text{s}$  flashlets of 30 000  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ , with a 1  $\mu\text{s}$  interval between adjacent flashlets, whilst an FRR MT saturation phase is formed of 3200 flashlets of 1  $\mu\text{s}$  duration with 49  $\mu\text{s}$  between flashlets. The sensitivity of the photomultiplier tube and the output from the excitation LEDs is monitored at each flashlet, to minimise measurement artefacts. Implementation of the ST FRR method provides information about PS II, over and above that provided by all other methods. Specifically, the level of connectivity among PS II complexes ( $p$ ), and the 'effective' absorption cross section of PS II ( $\sigma_{\text{PS II}}$ ) which is required for calculation of the electron transfer rate through each PS II complex ( $\text{ETR}_{\text{PS II}}$ ).  $\text{ETR}_{\text{PS II}}$  is often used in estimations of primary productivity.

## SAMPLE DATA

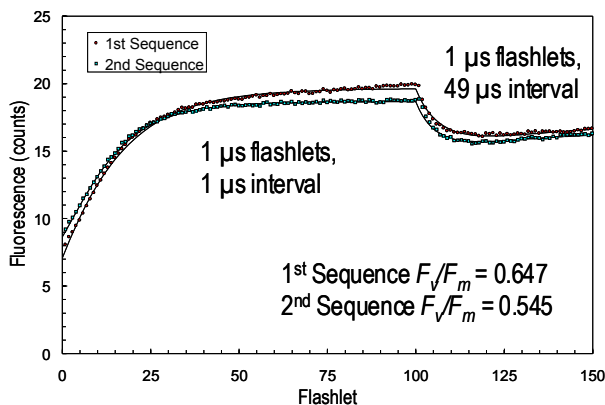


Data fit to the mean of sequences 2 to 6 of a 6 sequence acquisition from dark-adapted material. A mechanistic model (currently under development), which incorporates the turnover of plastoquinone, semi-plastoquinone and plastoquinol at the  $Q_B$  site of PS II, was used to fit these data.

The interval between flashlets during the relaxation phase started at 50  $\mu$ s, but was increased by 20% between consecutive flashlets. The model has generated a reasonable fit to the data, including the 'kink' in the relaxation phase.

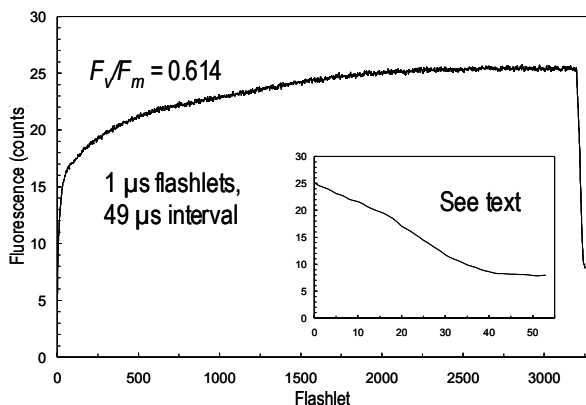


Same as above except that the interval between flashlets during the relaxation phase was fixed at 199  $\mu$ s. The mechanistic model has produced a reasonable fit to the data, including to the slow fluorescence increase during the latter part of the relaxation phase.



Analysis of individual sequences within an acquisition. To generate these data, six acquisitions of two sequences were made from dark-adapted cells, with a 30s intervals between acquisitions. The mean trace of the first sequences shows a lower value for  $F_0$ , but higher values for  $\sigma_{PS II}$  and  $F_m$  than for the second sequence.

The higher  $F_m$  value for the first sequence in an acquisition from dark-adapted material is a common feature of ST data. For example, Kolber et al. (1998) noted a 15 to 20% higher  $F_m$  after 10 s dark-adaptation.



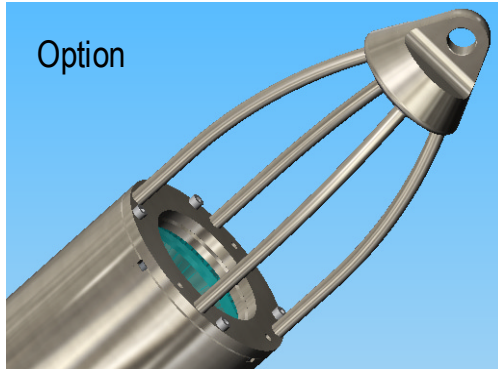
Plot of a single MT data sequence. The interval between flashlets during the relaxation phase started at 800  $\mu$ s, but was increased by 20% between consecutive flashlets.

The inset shows the relaxation phase in more detail.

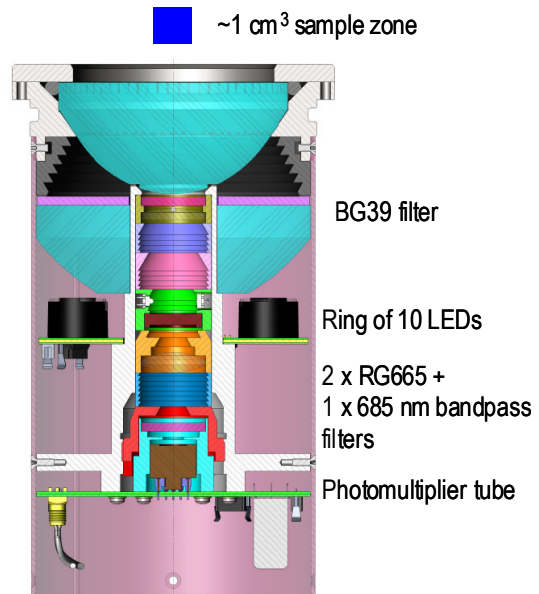
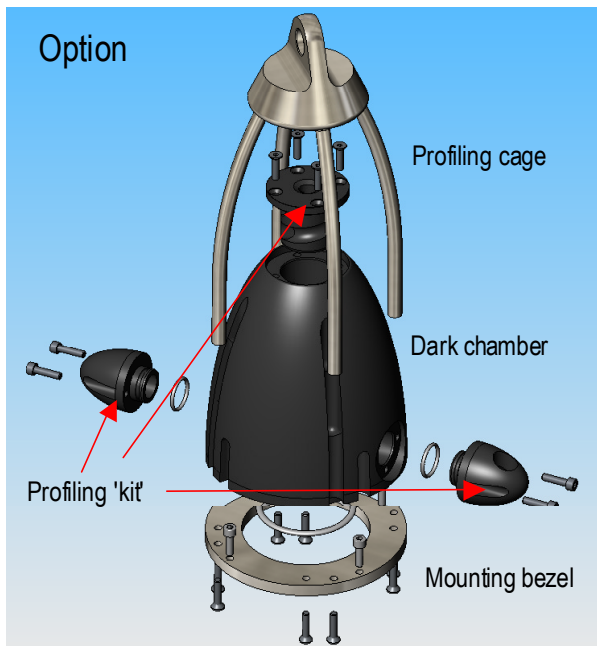
All of the examples shown above were collected from a mixed population of green algae, at a chlorophyll concentration of approximately 26  $\mu$ g l<sup>-1</sup> (Chl a in acetone equivalent). A series of 9 concatenated protocols was used to collect all of the above data in one go.

Reference: Kolber ZS, Prášil O and Falkowski PG (1998) Measurements of variable chlorophyll fluorescence using fast repetition rate techniques: defining methodology and experimental protocols. Biochim. Biophys. Acta 1367: 88-106

## SPECIFICATIONS and OPTIONS



The Fasttrack II profiling cage provides a high level of protection for the photomultiplier tube, against stray light.



The **Fasttrack II electro-optics assembly** provides very even illumination within the sample zone. The filtering and optical arrangement of the LEDs and photomultiplier tube, which excite and detect Chl *a* fluorescence, respectively, minimise contamination by stray light and 'filter breakthrough' from the LEDs.

The **Fasttrack II dark chamber** can be combined with the **profiling cage**, with the **bezel** providing the point of attachment to the sensor for both items. The **profiling adaptor kit**, shown here, provides good flow through when the sensor is used within a towed vehicle, for example. If the sensor is being used in a static location, or in slow moving water, a **pumped adaptor kit** (not shown) can be used with the same dark chamber.

### PHYSICAL

Sensor length	339.5 mm (ex. connector)
With profiling cage	550.0 mm (ex. connector)
Sensor diameter Ti (Ac)*	112.5 mm (130 mm)
With profiling kit	170.5 mm
Pressure housing	Titanium or Acetal C
Dark chamber	Acetal C
Connector kits	Acetal C + Stainless Steel
Subconn® connector	Stainless Steel
Weight in air / water	6kg / 3kg approx.

\*Ti = titanium pressure housing, Ac = Acetal C pressure housing

### OPERATIONAL

Range (standard)	0 - 600 µg l <sup>-1</sup>
Excitation	470 nm peak, ± 20 nm
Detection	685 nm peak, ± 10 nm
Sample volume	1 cm <sup>3</sup>
Internal storage	2 GBytes
Power in	9 to 72 VDC
Operating temperature	-10°C to +40°C
Storage temperature	-10°C to +50°C
Max. operating depth Ti (Ac)*	500 m (200 m)

**Basic instrument:** The Fasttrack II is provided with a 3 m cable, Handbook and Calibration Certificate and fitting software

**Available options:** Deck unit, profiling cage, dark chamber, profiling adaptor kit and pumped adaptor kit. PAR sensor and depth sensor



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