Rapid Kinetic and Spectroscopy instruments

SFM-100 and MOS LED: installation and system description in double ABSORBANCE Mode (Manual stopped flow)

- MOS LED Rear panel: (connect the power cord)
- MOS LED Front panel (cf figure 1 below):

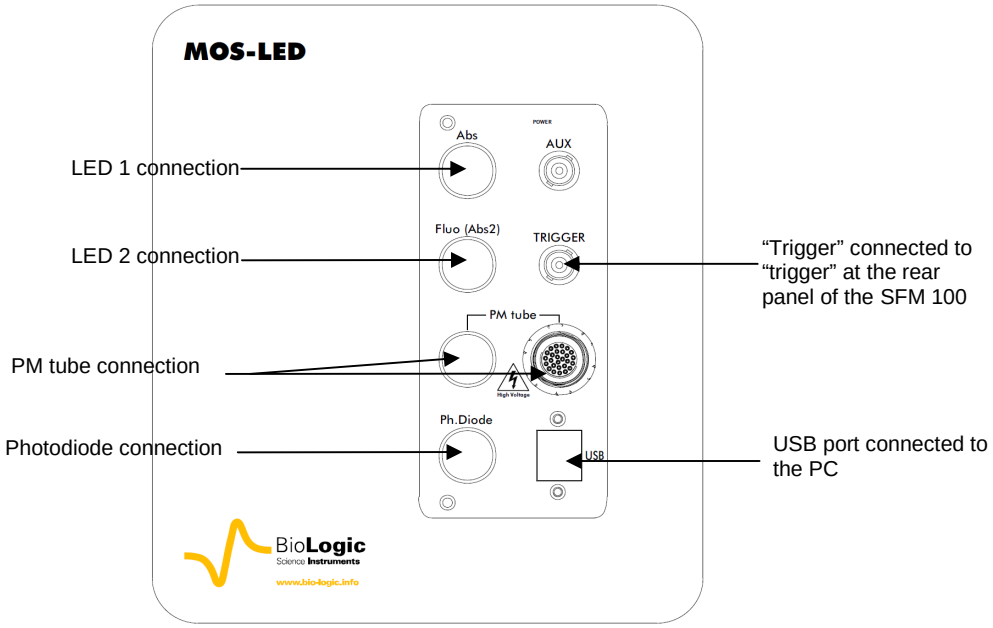

Figure 1: MOS- LED front panel connections

- SFM–100: rear panel connections

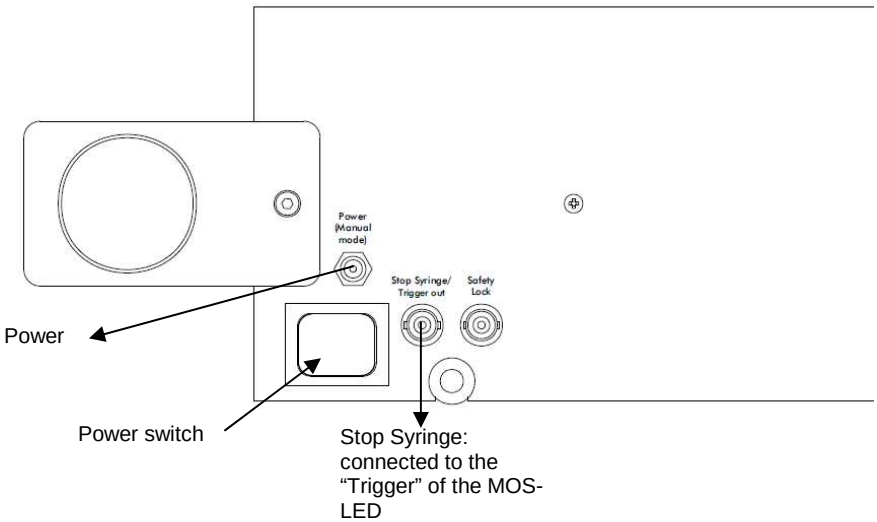

Figure 2: SFM-100 rear panel connections

SFM-100 description

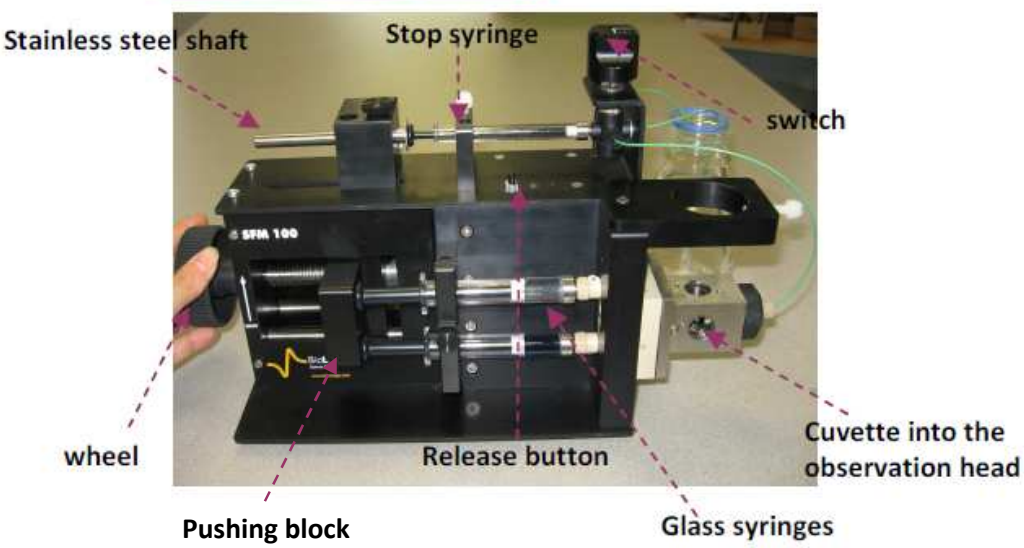

Figure 3: stopped flow description

In dual absorbance mode: the photodiode is placed at 180° of the light source 1(LED1) and the PM tube is placed at 180° of the light source 2 (LED 2) (cf figure below)

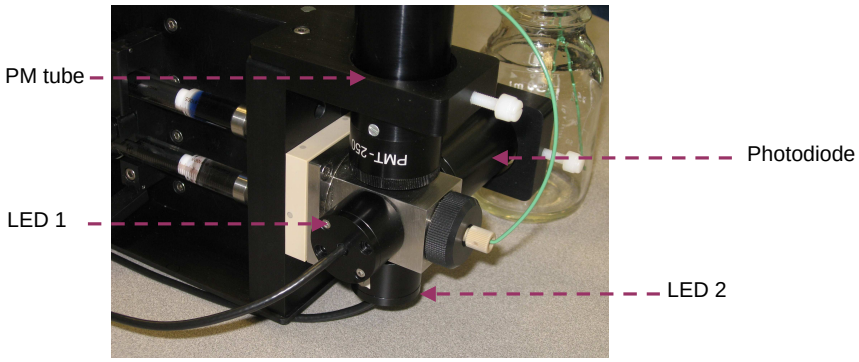

Figure 4: dual absorbance configuration

Double wavelength kinetics experiment

The aim of this application note is to learn how to perform a dual absorbance kinetics using the MOS-LED, SFM-100 and Spec-Lab.
Spec-Lab software should be installed and well configured.
The reduction of 2,6-dichlorophenolindophenol (DCIP) by L Sodium ascorbate is presented in this application note. The kinetic is followed by absorbance at 524 nm. If the concentration of DCIP is sufficiently smaller than Sodium Ascorbate then the reaction is treated as a pseudo first-order reaction.

Experimental conditions:

- **MOS-LED** : use of two LEDs: monochromatic light source peaking at 524 nm
  - Mode : absorbance
  - Detector: photodiode and PM tube

.

- **SFM-100** equipped with two syringes of 10ml (ratio 1:1)
  - Syringe 1 : 5 mM L- sodium ascorbate
  - Syringe 2: DCIP
  - Cuvette: µTC-100/10F (cuvette already installed)
  - Total flow rate : controlled manually with the driving wheel

Optical settings:

- Install the Photodiode at 180° to the LED 1 (on th e 1mm pathlength) , install the PM tube at 180° of the LED 2 to get a measurement on the 1 cm pathlength of the cuvette (cf scheme below of the µTC-100/10 cuvette)

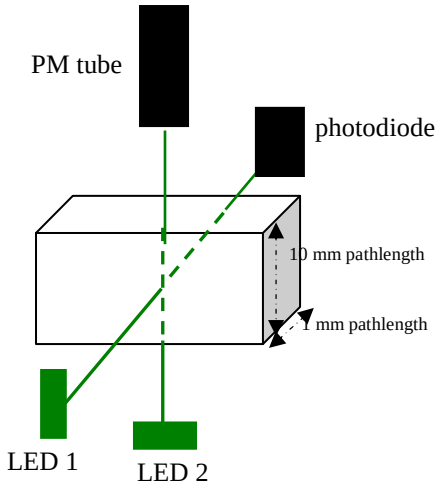

Figure 5: Cuvette position (µTC-100/10 F) and optical settings

Switch on the MOS-LED and SFM-100
Launch Spec-Lab software

- Click on “device” , and select MOS-LED and click on “connect”
- Click on technique and select “DABS” for dual absorbance measurements. A new window should arise ((figure 6)

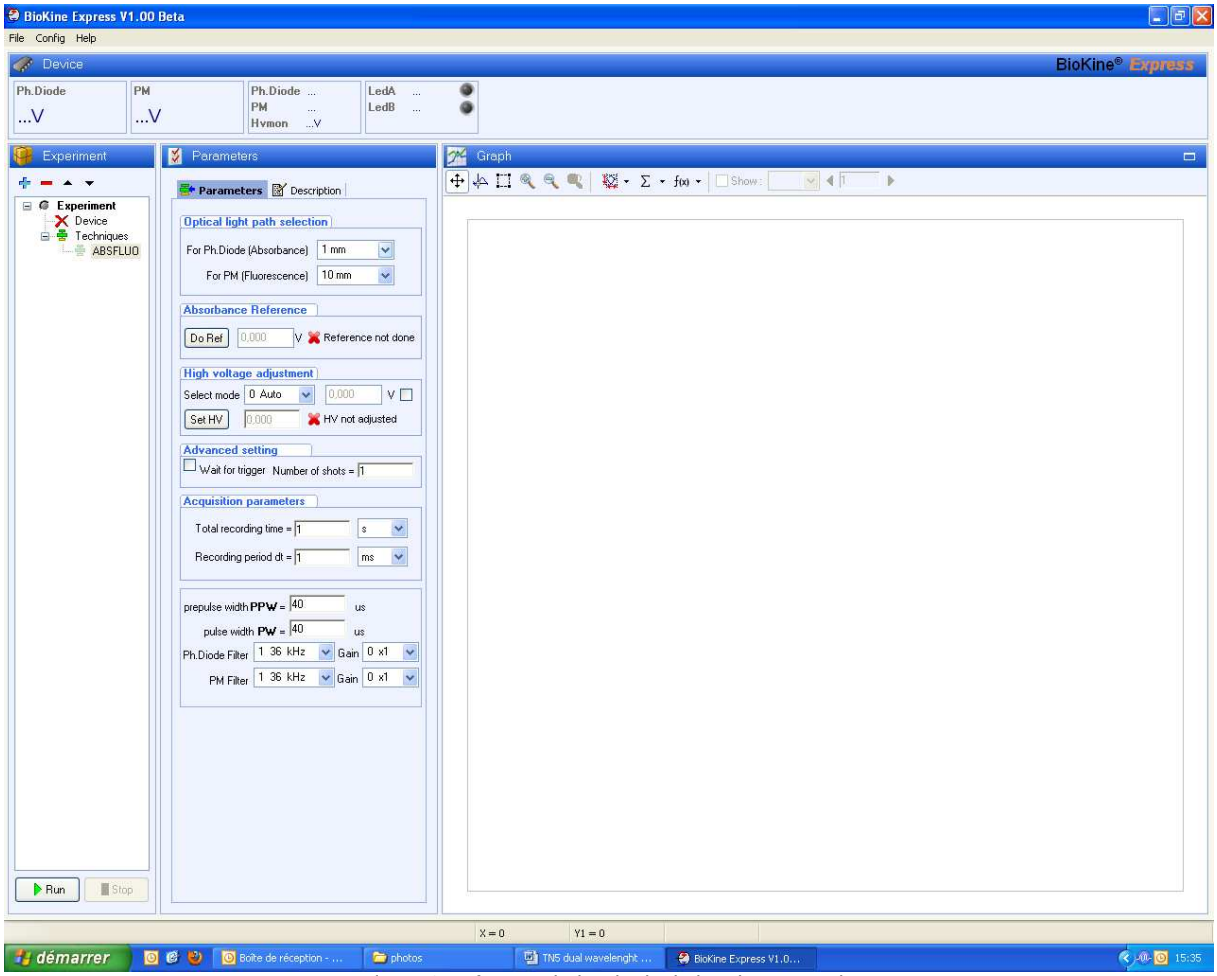

Figure 6: software window in dual absorbance mode

1/ First step is to fill the cuvette with water to do the reference. Use the wheel to move back the pushing block and unscrew the glass syringes. Once the syringes are filled with water, make sure to remove all air bubbles. Place back the syringes in the SFM, turn the switch of the SFM to the waste position (switch on ⏚) and turn the wheel following the arrow until the solutions flow from both syringes (i.e. pushing block must be in contact with the piston of the syringes): this procedure is called **the purge.**

2/ second step is to define the acquisition parameters in the software and do the reference.
- Choose the “optical light pathlength for photodiode“ : 1 mm
- choose the “optical light pathlength for PM tube”: 10mm
- Select the “wait for trigger” and “number of shot:1”
- Enter “total record time”: 4 s
- Enter “recording period”: 1 ms
- At this step water is in the cuvette, click on “DO REF” to do the blank: (Cf figure 7). Two voltages are displayed one around 5V and a HV over 100V.

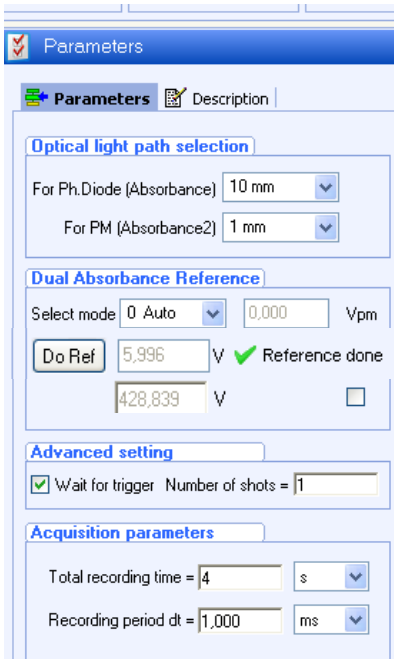

Figure 7: acquisition parameters

3/ Third step is to replace the water of the syringes against the samples DCIP and sodium ascorbate and then purge the system. Turn the switch clockwise to connect the cell to the stop syringe (position ◵). Press the on the release button of the SFM-100 (green LED brights). On Spec-Lab software, click on “RUN”. Enter the name of the file, and finally, turn the wheel of the SFM following the arrow to initiate the shot.
The kinetic is displayed in the window (figure 8): the first kinetic (blue) is recorded with the photodiode and the red kinetic is recorded with the PM tube.

4/ To do an other shot, empty the stop syringe: turn clockwise the switch to connect the stop syringe to the waste (position◴), push the stainless steal shaft. Turn clockwise the switch to the shot position ◵ and press on the release button. Click on “RUN” in the software and initiate the shot with the wheel.

Remark: to release the pressure from the stopped flow between each shot, we must reach this switch position◷: that’s why it is very important to turn clockwise the switch button

5/ results analyses:
To calculate the constant rates: click on “f(x)”, “multiexponential fit”, a new window opens; select the curve that has to be fitted, and click on “minimize” and then click on “calculate”, the constant rate k is then displayed

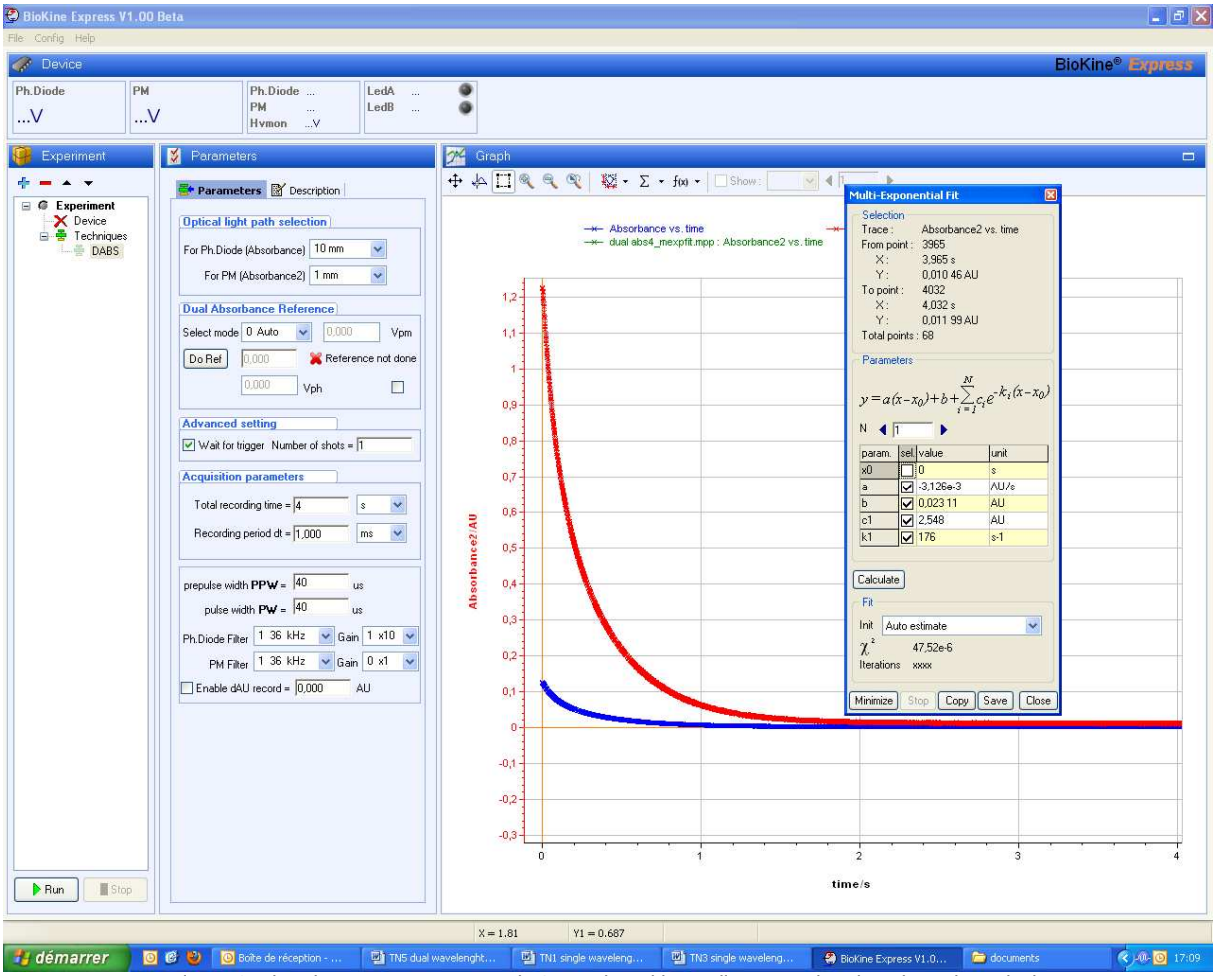

Figure 8: absorbance measurement (DCIP reduced by sodium ascorbate) and result analysis

For more information, please refer to the user manual or contact us through our website:
www.bio-logic.info