

## Fluorescence Lifetime Imaging (FLIM)

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Fluorescence Lifetime Imaging (FLIM) is the measurement of the lifetime of a fluorophore in an excited electronic state as a function of position on an image. This gives a picture of environment of the fluorophore. The lifetime of a fluorophore can change with chemical composition (pH, calcium concentration) or because of quenching (FRET, oxygen). At the SSSC, FLIM is based on the time domain acquisition of information in conjunction with the laser scanning confocal microscope. The pulsed laser system can be used in pico- or femtosecond mode with a variety of repetition rates, and it can be used in the visible or infrared regions.

#### **Specifications**

Excitation wavelength	Two-phonon mode: 700 to 1000 nm Visible: 400 to 490 nm
Objective lens	63x/0.9NA, 2.2 mm working distance
Detection	Descanned (2 detectors with emission filter wheel) for visible light excitation Non-descanned PMC-100-4 (Becker and Hickl) FWHM 190 ps
Acquisition	Data acquisition board SPC-830 (Becker and Hickl)
Advanced capabilities	63x/0.9NA, 2.2 mm working distance
Detection	MCP-PMT, R3809U-51 (Hamamatsu) FWHM 35 ps 160 to 190 nm
Acquisition	Data acquisition board: SPC-830 (Becker and Hickl) SPC 630 (Becker and Hickl)
Advanced capabilities	Objective inverter (LSM Technologies) for upright microscopy applications FCS2 Temperature control chamber (contact Dr. Jim Xiang for use of the FCS2) Detector: Cooled MCP-PMT R3809U-51 (Hamamatsu) with FWHM ~ 35 picosecond

#### **Settings**

This system uses the Verdi / Mira / Pulse Picker / Harmonic Generator suite.