

Fluorescence Lifetime Imaging Module

- Modular upgrade for existing microscopes
- Time-gated FLIM approach
- Lifetime discrimination <100ps

Description **Technical Description** Citations

Technical Description

Exemplar FLIM instruments

Wide-field time-gated fluorescence lifetime imaging microscope

The schematic below shows an overview of the components used for wide-field time-gated FLIM comprising the core elements : (i) *ultrafast laser excitation source*, (ii) *Gated Optical Intensifier (GOI)* and *computer-controlled delay generator*, (iii) *computer* for data acquisition and analysis, and (iv) a *wide-field epifluorescence microscope* with cooled readout camera.

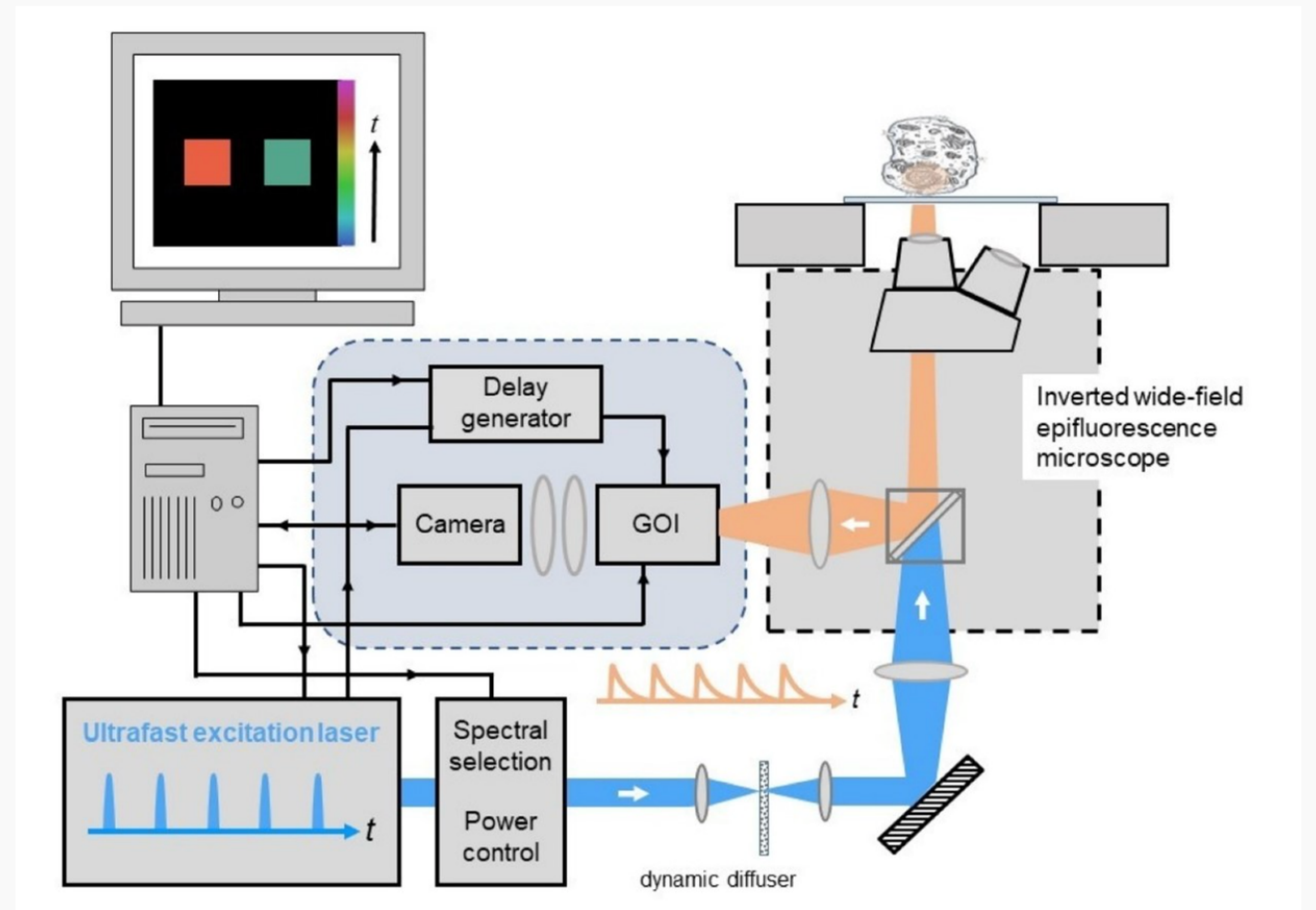


image: Wide-field time-gated FLIM

Wide-field optically sectioned time-gated fluorescence lifetime imaging microscope

For optically sectioned FLIM, the wide-field time-gated FLIM module can be integrated with optical sectioning using a Nipkow spinning disc scanner to provide **quasi-confocal FLIM** at frame rates up to 10Hz. This is particularly useful for quantitative live cell FLIM since the high degree of parallelisation (e.g. compared to laser scanning microscopy) greatly reduces the instantaneous light dose at the sample and so reduces photobleaching and phototoxicity. The instrument configuration is similar to that for widefield time-gated FLIM except that the excitation light is coupled to the spinning disc scanner via a polarisation-preserving single mode optical fibre, as depicted below, and the FLIM module is mounted at the camera port of the Nipkow disc scanning unit.

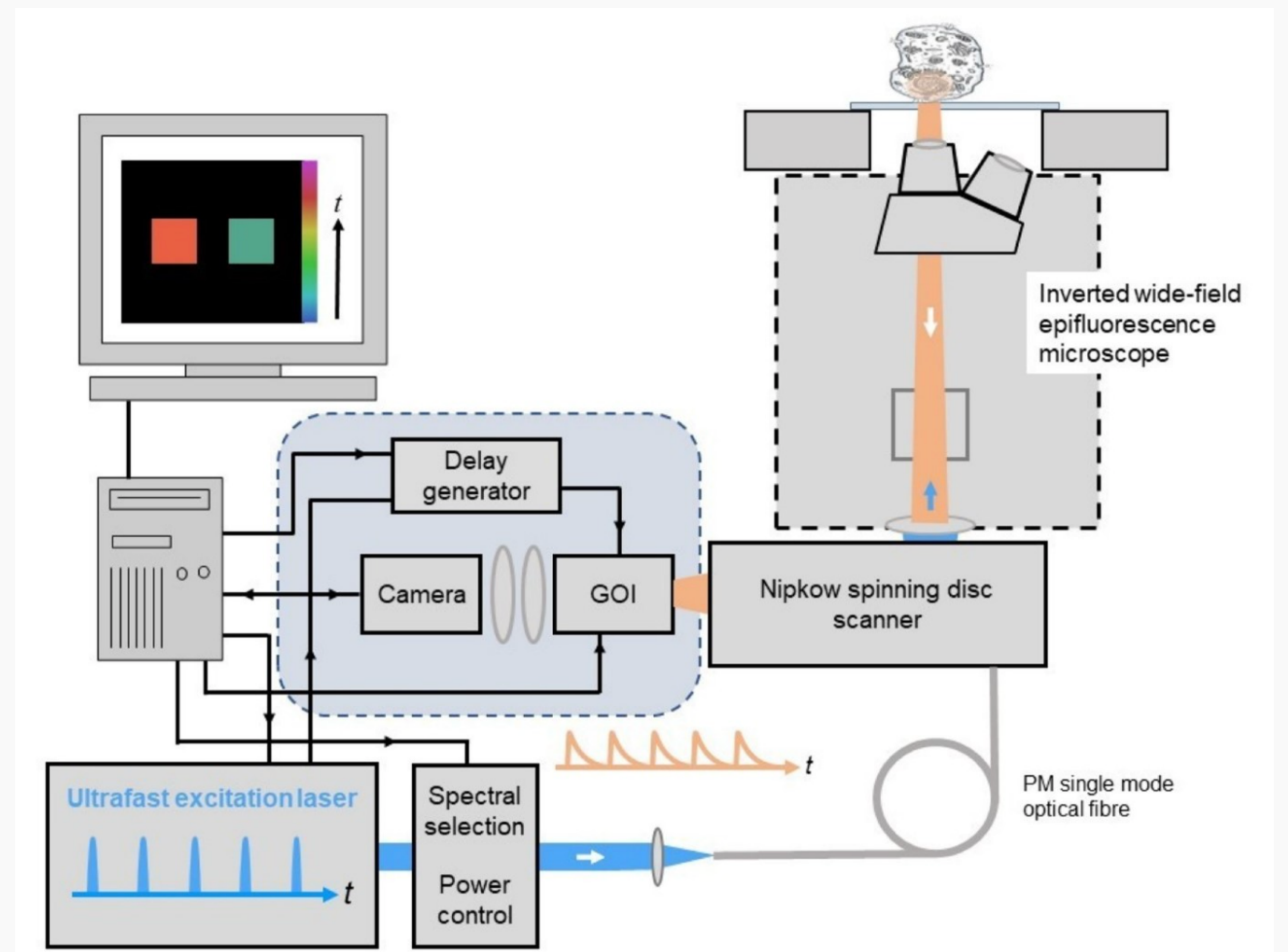


image: Optical sectioned time-gated FLIM

Both of these approaches can be implemented on fully motorised microscopes to provide automated multiwell plate imaging for **FLIM high content analysis**. Wide-field time-gated FLIM can also be combined with hyperspectral imaging to implement fluorescence lifetime resolved hyperspectral imaging and combined with polarisation-resolved imaging to realise time-resolved anisotropy imaging.

Wide-field time-gated FLIM has been applied to study protein interactions in cell-based studies and to tissue autofluorescence to explore label-free diagnosis of disease, including cancer.

openFLIM software

The system is configured with FLIM data acquisition achieved using the open source software, **openFLIM-GOI**, which is a **MicroManager** plug-in. This enables calibration and configuration of the system for specific experiments and saves the time-gated FLIM data as OME.TIFF. **openFLIM-GOI** was developed at Imperial College London.

FLIM data analysis may be undertaken using **FLIMfit**, an open source MATLAB-based software tool that provides an unrivalled range of fitting techniques including global fitting of regions of interest and global analysis.

QUESTIONS?

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ABOUT US

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