## STED/FLIM/FCS – SP8 (August 2020)

## **Technical Specifications**

- Leica SP8 AOBS confocal laser scanning microscope attached to a Leica DMi8 inverted epifluorescence microscope with 'Adaptive Focus Control' to correct focus drift during time-courses.
- Conventional scanner (100-1800 Hz) and resonant scanner (8 kHz) enable a broad range of applications including imaging at up to 25 frames per second with resonant scanner.
- Two standard PMTs plus two gated 'hybrid' SMD GaAsP detectors, which offer greater sensitivity, low noise and photon counting modes as well as time gating to maximise the signal to noise of measurements.
- Transmitted light detector.
- Spectrophometers allow customised detection of emitted light, spectral scanning etc.
- Equipped with 120 mW 405 nm diode, 75 mW 442 nm and white light laser (WLL) 470 -670 nm with 2.5 mW/nm and pulse picker to select 80,40,20 or 10 MHz rep rates.
- Combining spectral detection with WLL enables excitation/emission scanning (lambda<sup>2</sup>).
- Two STED lasers 592 nm and 660 nm provide depletion for green and red fluorophores.
- Picoquant electronics allow TCSPC fluorescence lifetime data acquisition and fluorescence (cross) correlation spectroscopy measurements.
- AOTFs for all laser lines allow rapid attenuation and switching between (up to 8) WLL lines.
- AOBS (Acousto-Optical Beam Splitter) automatically adjusts to selectively reflect each excitation line and allows optimisation of detection close to (and overlapping) excitation lines.
- Notch filters block reflection signal, enabling clearer imaging close to coverslips.
- Suitable for a wide range of blue, cyan, green, red and far-red fluorophores.
- Environmental chamber (Life Imaging Services) for temperature and CO<sub>2</sub> enrichment.
- Märzhäuser scanning stage enables multi-position acquisition and tiled imaging.
- HyVolution 2 improved resolution (to ~140nm) with specimens prepared for standard confocal imaging.

Position	Excitation range	Fluorophore (examples)			Emission filter	Serial Number
1	UV	DAPI	350/50	400	460/50	525301
2	Blue	FITC	480/40	505	527/30	525302
5	Green	Rhodamine	546/40	560	585/40	525303

## Filters for visual inspection

## Default Lenses

Lens	Dry/ Oil	Phase contrast	DIC	Working distance (mm)	Numerical aperture	Features	Serial number	Image size at 0.75x zoom in microns
10x HC PL Fluotar	Dry	No	No	11	0.3		506507	1550
20x HC PL APO CS2	Dry	No	Yes	0.62	0.75		506517	775
40x HC PL APO CS2	Oil	No	Yes	0.24	1.3		506359	387.5
63x HC PL APO CS2	Water	Yes	Yes	0.65	1.2	Motorised correction collar	506361	246
63x HC PL APO CS2	Oil	Yes	Yes	0.14	1.4		506351	246

Lens	Dry/ Oil	Phase contrast	DIC	Working distance (mm)	Numerical aperture	Features	Serial number	Image size at 0.75x zoom in microns
93x HC PL APO CS2	Gly	No	Yes	0.3	1.3	Motorised correction collar	506417	166.667
100x HC PL APO CS2	Oil	No	Yes	0.09	1.4		506378	155