

# Luminosa **NEW**

## Single Photon Counting Confocal Microscope

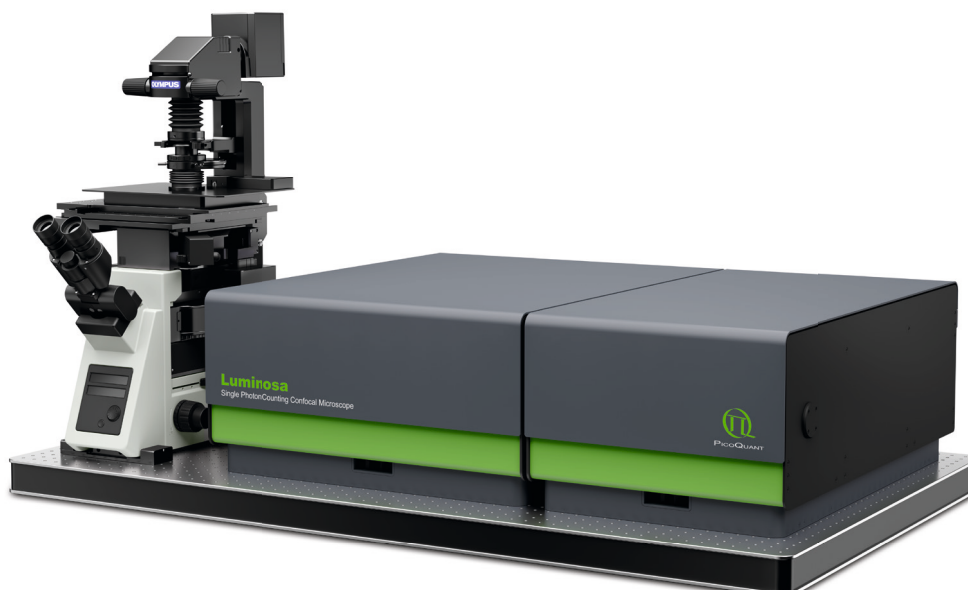
- Software-controlled confocal system based on an inverted microscope
- Versatile excitation system with laser wavelengths from 375 to 1064 nm
- VarPSF: fine-tuned observation volume for FCS and single molecule FRET experiments
- Motorized positioning table for “tiling and stitching” in transmission and FLIM mode
- Scanning options: FLIMBee galvo scanner and piezo objective scanning
- Up to 6 truly parallel detection channels with SPAD and/or Hybrid PMTs
- < 700 ps dead time per channel and 5 ps time bins
- One click autoalignment for consistent optimal performance
- Fast results with minimal user interaction thanks to GPU accelerated algorithms and context-based workflows for FCS, FLIM, and single molecule detection
- Imaging Scanning Microscopy (ISM-FLIM): FLIM with more spatial resolution and higher contrast

### Core methodologies

- Single molecule FRET (burst and time trace analysis)
- Fluorescence Lifetime Imaging (FLIM)
- Fluorescence Correlation Spectroscopy (FCS)
- FLIM-FRET
- rapidFLIM<sup>HiRes</sup> for fast processes
- Anisotropy Imaging
- DIC imaging
- Imaging Scanning Microscopy (ISM-FLIM)

### Application areas

- Dynamic structural biology
- Studying cellular mechanisms driven by phase separation
- Environmental sensing and marker multiplexing
- Mapping dynamics and structure of cellular membranes
- Characterizing functional nanovesicles
- Studying chemical reactions at the single molecule level
- Characterizing advanced materials



## Specifications

Feature highlights	
LumiFinder	Automated detection and measurement of immobilised emitters
FRETcompass	Automated determination of correction factors used in single molecule FRET experiments based on the benchmark study published in 2018 (Hellenkamp et al. Precision and accuracy of single-molecule FRET measurements—a multi-laboratory benchmark study. Nat Methods 15, 669–676 (2018))
InstaFCS	Online FCS fitting with automatic suggestions for model and parameters
InstaFLIM	<ul style="list-style-type: none"> <li>Provides first suggestions for FLIM species separation without user interaction</li> <li>Suggested parameters can be fine-tuned</li> <li>Simultaneous TCSPC and phasor analysis options for ROI determination</li> </ul>
Sample-Free Auto-alignment (SFA)	<ul style="list-style-type: none"> <li>One-click auto-alignment without any sample required</li> <li>Ensures optimal performance for every measurement, even the most challenging ones</li> </ul>
CalEx*	<ul style="list-style-type: none"> <li>Excitation laser power calibration allows setting and displaying excitation intensity in <math>\mu\text{W}</math></li> <li>No external power meter required</li> </ul>
VarPSF*	Switch between diffraction limited and larger observation volume to: <ul style="list-style-type: none"> <li>Increase observation time window to check for dynamic transitions between states</li> <li>Study diffusion properties of larger particles, 3-6 times larger volumes are achieved (depending on wavelength)</li> </ul>
Luminosa Full-Access Mode (LFA)	Fully accessible optical unit with dichroics and filters that can be easily exchanged, no special tools required
Excitation	
	1-8 lasers with choice of wavelengths from 375 - 1064 nm optional: 2 additional exit ports available for coupling additional lasers
Commonly used wavelengths	405, 440, 485, 532, 560, 595, and 640 nm
Individual laser operation modes	pulsed and continuous wave (continuous mode availability depends on the laser model)
Repetition rates	1 - 40 MHz, for specific wavelengths up to 80 MHz
Laser driver	8 channel Sepia PDL 828L
Supported multi line operation	Pulsed Interleaved Excitation (PIE) or simultaneous emission modes (for specific wavelengths Alternating Excitation mode (ALEX) in $\mu\text{s}$ and ms time scales also available)
Motorized main dichroic wheel	6 positions available for $18 \times 25 \times 3$ mm single, dual, triple, quad-band dichroics
Microscope	
	inverted Olympus IX73 body (partially motorized)
Objectives types	UPLSAPO 60x PlanApochromat, NA 1.2, water immersion, 400 - 900 nm UPLSAPO 100x PlanApochromat, NA 1.4, oil immersion, 400 - 850 nm other oil immersion, apochromatic correction, air spaced, IR-enhanced or long working distance objectives available
Observation modes	confocal, transmission optional: epi-widefield illumination, Differential Interferometric Contrast (DIC)
Oculars	Yes
Camera	Optional CCD camera at the left side port

Sample holder	Multi-functional sample holder compatible with several standard sizes of microscope coverslips and petri dishes 96-well plate sample holder	
Compatible with cage incubator	Yes	
Positional stability		
Axial drift (typical values)	<ul style="list-style-type: none"><li>For 15 minutes measurement: 50 ± 10 nm</li><li>For 2 hour measurement: 100 ± 20 nm</li></ul>	
Point Detection Option		
	<ul style="list-style-type: none"><li>Parallel detection with up to 6 point detectors in the spectral range of 300 - 1064 nm</li><li>Additional exit ports for customised detection path</li></ul>	
Point Detector Type		
	<ul style="list-style-type: none"><li>Single Photon Avalanche Diodes (SPAD) for low dark-counts and maximum detection efficiency</li><li>Hybrid photomultiplier tubes (PMA-Hybrid-40) for optimum timing performance and when working at high count rates</li><li>23 pixel SPAD array</li></ul>	
Type	SPAD (SPCM-AQRH)	PMA Hybrid - 40
Spectral range	400 - 1000 nm	300 - 720 nm
Dark counts (at 20 °C, typ. value)	< 250 cps	< 300 cps
Photon detection efficiency	50 % at 550 nm	40 % at 500 nm
Timing response	200 - 400 ps (depending on wavelength)	100 ps
Spectral separation	Fluorescence filters (25 mm in diameter) and 25 × 36 × 1mm dichroics	
Polarisation separation (optional for anisotropy imaging)	Polarising beamsplitting cube	
PDA-23 Array Detector of 23 independent Single Photon Avalanche Diode pixels		
Active area	100 µm x 110 µm	
Effective fill factor with microlenses	> 75% , typical 80%	
Photon detection efficiency (PDE)	Typ. Peak of PDE 55% at 520 nm PDE typically above 35% for wavelengths from 440 to 660 nm Usable spectral window 400 -950 nm	
Dark counts at working temperature of 10 Degree via integrated cooling	Typ. 100 cps for most pixels Median value of Dark counts per second is < 100. Typ. 1 hot pixel with > 1 kcps and below < 10kcps Hot Pixel not in the center of the array.	
Jitter	< 100 ps FWHM for wavelengths between 510 and 785 nm	
Deadtime of each pixel	50 ns	
Cross talk probability between pixels	0.1%	
Afterpulsing probability	0.1%	
Maximum Count rate per pixel	Typically > 7 Mcounts per second	

Data acquisition	
	<ul style="list-style-type: none"> <li>based on Time-Correlated Single Photon Counting (TCSPC) in Time-Tagged Time Resolved (TTTR) measurement mode</li> <li>simultaneous data acquisition with up to 6 channels</li> <li>new dynamic binning format for FLIM imaging</li> </ul>
<b>TCSPC Unit</b>	<b>MultiHarp 150 8P</b>
Time resolution (bin width)	5 ps
Dead time	< 0.65 ns
Timing precision	< 32 ps rms
Scanning and positioning	
FLIMbee XY galvo scanner	Min. pixel dwell time: 0.5 $\mu$ s, Min. pixel size: 17 nm (60x objective) Up to 5.2 frames per second for a 512 x 512 pixel image Max. scan field: 200 x 200 $\mu$ m (60x objective) Possibility to by-pass the FLIMbee galvo scanner for point measurements
XY objective scanning	Min. pixel dwell time: 0.2 ms Min. pixel size: 1 nm (independent of objective) Max. scan field: 80 x 80 $\mu$ m (independent of objective)
XY - scanning combined option	Combination of FLIMbee and objective scanning on the same system with software-controlled change between the two scanning modalities
Piezo-based Z-scanning	Min. step size: 50 nm Overall range: 100 $\mu$ m
Positioning stage with Spiral scan mode, creating overview map by tiling and stitching	Overall range: 121 x 81 mm Max speed: 300 mm/second Positioning repeatability < 0.15 $\mu$ m
Luminosa software	
Operating System	Windows 11
GPU – based programming	OpenGL
Context based workflows for easy acquisition and analysis	For single molecule FRET burst analysis, single molecule imaging, FCS in vitro, FCS in cells, FLIM, FLIM-FRET, steady-state FRET, anisotropy imaging
General info	
Operating environment	Room temperature range 15 - 25 °C Room temperature stability $\pm$ 1.5°C (recommended) Room humidity < 60%
Optical table dimensions	For 1-4 detectors: 1500 x 900 mm For 5-6 detectors: 2000 x 900 mm
Power consumption	6 A at 230 V AC (typ. EU) 20 A at 110V AC (typ. USA)
Operating voltage	115 or 230 V AC
Altitude	Guaranteed performance up to 2000 m above sea level

\* Feature available only for PicoQuant lasers



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