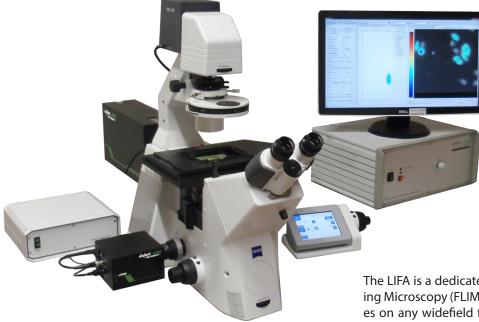


Fluorescence Lifetime Attachment

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The LIFA is a dedicated system for Fluorescence Lifetime Imaging Microscopy (FLIM). It allows the generation of lifetime images on any widefield fluorescence microscope using frequency domain technology. The LIFA model consists of a TRiCAM M modulated intensified CCD camera, a modulated lightsource, a modulation control unit, a personal computer and the LI-FLIM software package. The intensified camera contains an image intensifier that is fiber-optically coupled to the CCD camera for maximum gain and speed.

KEY FEATURES

Easy coupling

Flexible and efficient coupling to all major-brand fluorescence microscopes.

Fast acquisition Record lifetime images in a matter of seconds.

High resolution image intensifiers

Gen II and Gen III image intensifiers offering the world's highest resolution and sensitivity in the UV, visible or near infrared.

Accurate lifetime determination Fluorescence lifetimes from 0–300 ns with an accuracy of 30 ps r.m.s.

APPLICATIONS

Fluorescence Lifetime Imaging Microscopy (FLIM)

FRET Efficiency Mapping

Protein Interactions

Biosensors

Oxygen Concentration Imaging

NADH/FAD Fluorescence Dynamics

Membrane Dynamics

Molecular Interactions

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SPECIFICATIONS

| Specifications | Typical value |
|--------------------------|--|
| System Performance | |
| Lifetime range | 0 - 300 ns |
| Lifetime resolution | less than 100 ps (single pixel) |
| Speed | 2 lifetime images per second (full frame) |
| Spatial resolution | 26 lp/mm (Gen II); 19 lp/mm (Gen III) |
| Sensitivity | single photon |
| Intrascene dynamic range | 12 bit |
| Camera input format | 1 inch |
| Effective pixel size | 10.3 μm square (12,9 for lens coupled CCD) |
| | |

CONTROL UNIT

The control unit is used to generate and control the modulation signals for the lightsource and the intensified camera. It also contains the high voltage power supply generating the DC voltages for the image intensifier. All variables are controlled in the LI-FLIM software via a USB 2.0 interface.

| Signal shape | Sinusoidal |
|--|--|
| Frequency range | 1 MHz – 120 MHz |
| Frequency adjustment resolution | 1 kHz |
| Frequency deviation | 100 ppm max. |
| Harmonics suppression | Better than 25 dB |
| Output 1 Type a: MultiLED AC signal level DC current adjust | Capable of driving lightsource type a, optionally b and c Adjustable in 250 steps 0 – 1000 mA, 12 bit resolution |
| Optional lightsource outputs: Type b: Modulated laser diode AC signal level DC level adjust Type c: AOM for CW laser AC signal level Frequency Output 2 AC signal level AC signal after amplification | 0.00 – 1.00 V peak-peak 0.00 – 1.00 V 0.00 – 1.00 V peak-peak Half the frequency of output 2 To image intensifier Adjustable from –50 to 0 dBm 15 Vpp nom. |
| External trigger input | LVTTL (phase shift on falling edge or rising edge) |
| Impedance | 50 Ω |
| Phase adjustment range | 0 - 360° in 1° steps |
| Phase deviation | < 0.01° |
| Cathode voltage DC | Adjustable from –150 V to +15 V |
| MCP voltage (gain) DC | Adjustable from 450 – 1000 V max. |
| Anode voltage DC | Fixed 6 kV |
| | |

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MULTI-LED LIGHT SOURCE

The Multi-LED is a modulated light source designed for fluorescence excitation. In the Multi-LED, up to four LED units are mounted having one common optical output to the microscope. Available LEDs cover the wavelength range from 360 to 660nm. The unit is cable-connected to the LIFA control unit that delivers the DC and AC power to the selected LED. LED selection, DC current, modulation frequency, amplitude and phase are controlled via the user interface of the LI-FLIM software. Optionally the Multi-LED can be temperature controlled for improved long-term stability. The Multi-LED is available for any brand of wide field microscope.

| Peak wavelengths (nm), approximately | 405 (CFP), 445 (cerulean, CFP), 470 (GFP), 535 (TRITC), 630 (CY5), others on request | |
|---|--|--|
| Number of mounted LEDs | 3 (standard), or 4 (maximum) | |
| Modulation frequency | up to at least 80 MHz | |
| LEDs switching time | 200ms | |
| Coupling | Compatible with existing Hg and Xe microscope adapters | |
| Lifetime drift | less than 30 ps* | |
| Optional Temperature stabilization for long-term (hours) lifetime stability | Active thermoelectric water cooling/heating system | |
| Control temperature range | 5 – 45° C | |
| Temperature stability (constant load, constant ambient temp) | ± 0.1° C | |
| Dimensions (I x w x h) | 346 x 109 x 183 mm | |
| * Measured using stable fluorescent material with single lifetime component of 3.2 ns | | |

MULTI-LASER LIGHT SOURCE

The modulated laser diodes of the Multi-LASER offer the high stability and high modulation depth required for high-accuracy lifetime imaging, and can also be used for regular fluorescence intensity imaging. The Multi-LASER uses a kineFLEX fiber delivery system to easily couple into your microscope stand. The Multi-LASER is a modulated light source for multi-beam confocal, TIRF, and widefield illumination.

Two versions are available: the standard version and the extreme version for even better power stability and less lifetime drift.

| Wavelengths (nm) | 375 (BFP), 405 (CFP), 445 (cerulean, CFP), 488 (GFP), 515 (YFP, Venus), 647 (CY5), others on request | | |
|------------------------------|--|--|--|
| Number of laser lines | Standard: up to 3 or up to 6; Extreme: up to 4. All combined into one optical fiber output | | |
| Output power (mW) | 20 mW-300 mW, depending on the wavelength | | |
| Modulation frequency | Up to 120 MHz (standard) | | |
| Input signal | Digital: 01 V / 50 Ω | | |
| Control interface | RS232 | | |
| Coupling | KineFLEX system, with a single-mode polarization maintaining fiber and either FC-APC, FC-PC or FC-P8 output connector | | |
| Power stability | Standard: 2%*, Extreme: 0.5%* | | |
| Lifetime drift | Standard: less than 50 ps*, Extreme: less than 20-30 ps* | | |
| Dimensions (I x w x h) | 19" rack housing. MultiLASER-3: 480 x 484 x 88 mm; MultiLASER-4 and 6: 610 x 484 x 132 mm | | |
| Options | Multi-channel excitation Two single-mode optical fiber outputs Custom fiber outputs on special request | | |
| * Measured using stable fluc | * Measured using stable fluorescent material with single lifetime component of 3.2 ns | | |

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TRICAM M INTENSIFIED CCD CAMERA

The TRiCAM M is the modulated version of the TRiCAM modulated intensified camera manufactured by Lambert Instruments. The TRiCAM is a compact camera based on a 18 mm MCP image intensifier that can be gain-modulated at high frequencies to allow frequency-domain imaging. The intensifier can also be operated DC to allow focusing and selecting the object of interest. Optionally, the TRiCAM can be provided with gating to allow time-gated imaging. AC and DC voltages are provided from the control unit via a cable that is permanently connected to the camera for maximum performance. The output of the intensifier can be either fiber-optically or lens coupled to the high-resolution progressive scan CCD camera with 12 bit output. The image acquisition and camera parameters as exposure time, binning, ROI, triggering are controlled via the USB2.0 interface.

| Resolution on input | Minimum | Typical | Units |
|---|--|------------------------------|-----------------------|
| Intensifier tube at nominal DC operation | Gen II: 55 | Gen II: 65 Gen III: 57 | lp/mm |
| Including relay lens or fiber optics, at DC operation and CCD camera | | Gen II: 40 Gen III: 34 | lp/mm |
| Including relay lens or fiber optics, at 40 MHz gain modulation and CCD camera | | Gen II: 26 Gen III: 19 | lp/mm |
| | | | |
| Image intensifier | Minimum | Maximum | Units |
| Input diameter | 17.5 | | mm |
| Input window | Borosilicate glass | | |
| Cathode sensitivity (Gen II: Super S25, Gen III: GaAs)* | | | |
| @ 2850 K @ 800 nm @ 850 nm | Gen II: 400, Gen III: 1000 35 25 | 1200 | μA/Im mA/W mA/W |
| Phosphor | P43 | | |
| Luminance gain | Gen ll: 3180 Gen lll: 3600 | | cd/m²/lx lm/m²/lx |
| Equivalent Background Input | | Gen ll: 0.25 Gen lll: 0.5 | μlx |
| Modulation frequency range | 1 | 120 | MHz |
| Quality area | Gen II: 14.4 x 10.7; Gen III: 13. | 5 x 10.0 | mm |
| | | | |

The number of spots exceeding a contrast with their surrounding area of 30% is less than or equal to the number indicated in the table below. The size of non-circular spots is determined on the basis of equal area to circular spots. When the distance between two spots is less than the maximum dimension of either spot, the two spots are considered to be one spot.

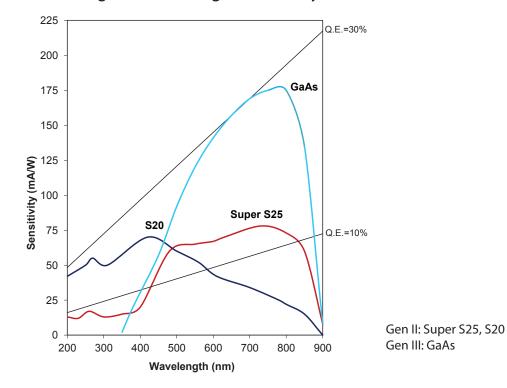
| Gen II | | Gen III | | |
|--|--|---|-----------------------------------|-----------------------------|
| Size of spots | Max. number of spots within effective area | Size of spots | Max. number of within effective | • |
| | | | Dark spots | White spots |
| > 300 μm 225 – 300 μm 150 – 225 μm 75 – 150 μm < 75 μm | 0 1 3 6 Minimal | > 150 μm 100 – 150 μm 75 – 100 μm 50 – 75 μm < 7 μm | 0 2 8 Minimal Minimal | 0 0 4 6 Minimal |
| | | | | |

* Photocathodes with different spectral response are available, see curve:



| CCD Camera | |
|---|--|
| CCD | 2/3" interline, progressive scan |
| Resolution | 1392(H) x 1040(V) pixels |
| Pixel size | 6.45 μm square |
| Dynamic range | 66 dB |
| Synchronized with | Control Unit |
| Integration time control | 1ms – 2 min |
| Selectable Region of Interest 1392 x 1040 1280 x 1024 1024 x 768 800 x 600 640 x 480 | @ 15 fps @ 16 fps @ 20 fps @ 24 fps @ 30 fps |
| Binning | 2 x 2, 3 x 3 and 4 x 4 |
| Fastest mode (4x4 binning = 348x256) | @ 50 fps |
| Digital output | Software selectable 8 or 12 bit, via USB |
| External trigger input | LVTTL |
| Trigger output | LVTTL |
| Pixel clock | 21 MHz |
| Programmable camera gain | Up to 23 x |
| High light level protection | Switches off all DC and AC power |
| Environmental temperature | 0° to 40° C |
| Lens mount | C-mount |
| Dimensions (I x w x h) | 133 x 116 x 80 mm |
| | |

Image intensifier light sensitivity



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PERSONAL COMPUTER (MINIMAL SPECIFICATION)

| Operating system | Windows 7 Professional 64-bit |
|--|--|
| Processor | Intel Core 2 Duo, 3GHz |
| Memory | 4GB |
| Hard disk | 300 GB |
| Monitor | 24" LCD monitor, resolution: 1920x1080 |
| System includes | DVD R, IEEE1394, LAN/Ethernet, USB2.0, keyboard, optical mouse |
| | |
| Software | |
| LI-FLIM | Acquisition and lifetime modules, full digital control of hardware, viewer and image analysis tools. External control via ActiveX. |
| Software Development Kit for CCD camera and Control Unit | Offers full programmatic (C/C++) control of camera and of Control Unit functionality. |

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