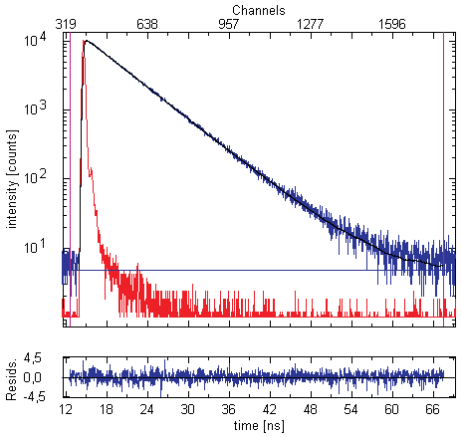
FluoTime 100

PicoQuant

Compact Fluorescence Lifetime Spectrometer



- Compact plug-and-measure design
- Based on Time-Correlated Single Photon Counting (TCSPC)
- Easy to use picosecond diode laser or sub-nanosecond LED light source
- Data acquisition in shortest time
- Filters for high optical throughput
- Advanced data analysis

Applications

- Time-resolved fluorescence spectroscopy and anisotropy
- Ultra sensitive analysis
- Photochemistry
- Marker and assay development
- Ideal for research and teaching purpose

Configuration and Standard Components

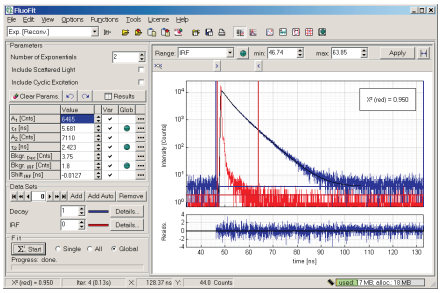
The FluoTime 100 is an easy to use, ultra compact time-resolved fluorescence spectrometer. It includes the complete optics and electronics for recording fluorescence decays via Time-Correlated Single Photon Counting (TCSPC). The system can be used with either picosecond diode lasers (LDH Series) or sub-nanosecond pulsed LEDs (PLS Series). With the FluoTime 100, decay times as low as 40 picoseconds can be resolved. The system allows operation at laser repetition rates as high as 85 MHz and count rates up to several million counts/sec.

Laser coupling module
The laser coupling module consists of a holder for uncooled laser/LED heads interlocked with the detector. The holder locks the orientation of the laser head to emit either vertically or horizontally polarized radiation. The beam is directed towards the sample by an adjustable steering mirror. A mounting plate for a cooled LDH Series head or a mirror holder is additionally available.

Sample chamber
The sample chamber contains a versatile sample holder for cuvettes. The tubing for a circulating fluid is pre-installed. A front face sample holder and a fiber probe are available as an option. Holders for filters, sheet polarizers and lenses in excitation and emission are positioned inside the sample chamber. For security, the sample chamber lid is interlocked with the detector.

Filter wheels
Two filter wheels are the components of the spectrometer, one for regulating the intensity of the excitation light, the other for filtering scattered excitation light and for selecting the spectral range of interest for detection. The standard excitation filter holder contains filters of 50 %, 10 %, 1 % and 0.1 % transmission. The emission filter wheel holds various cut-off and/or bandpass filters depending on the customer's request.

Data analysis
Numerical analysis of the raw data is an integral part of the TCSPC method. FluoFit is a Windows™ based decay analysis software with an easy to use graphical user interface and presentation-ready numerical and graphical output. It implements an iterative reconvolution fitting routine with nonlinear error minimization.

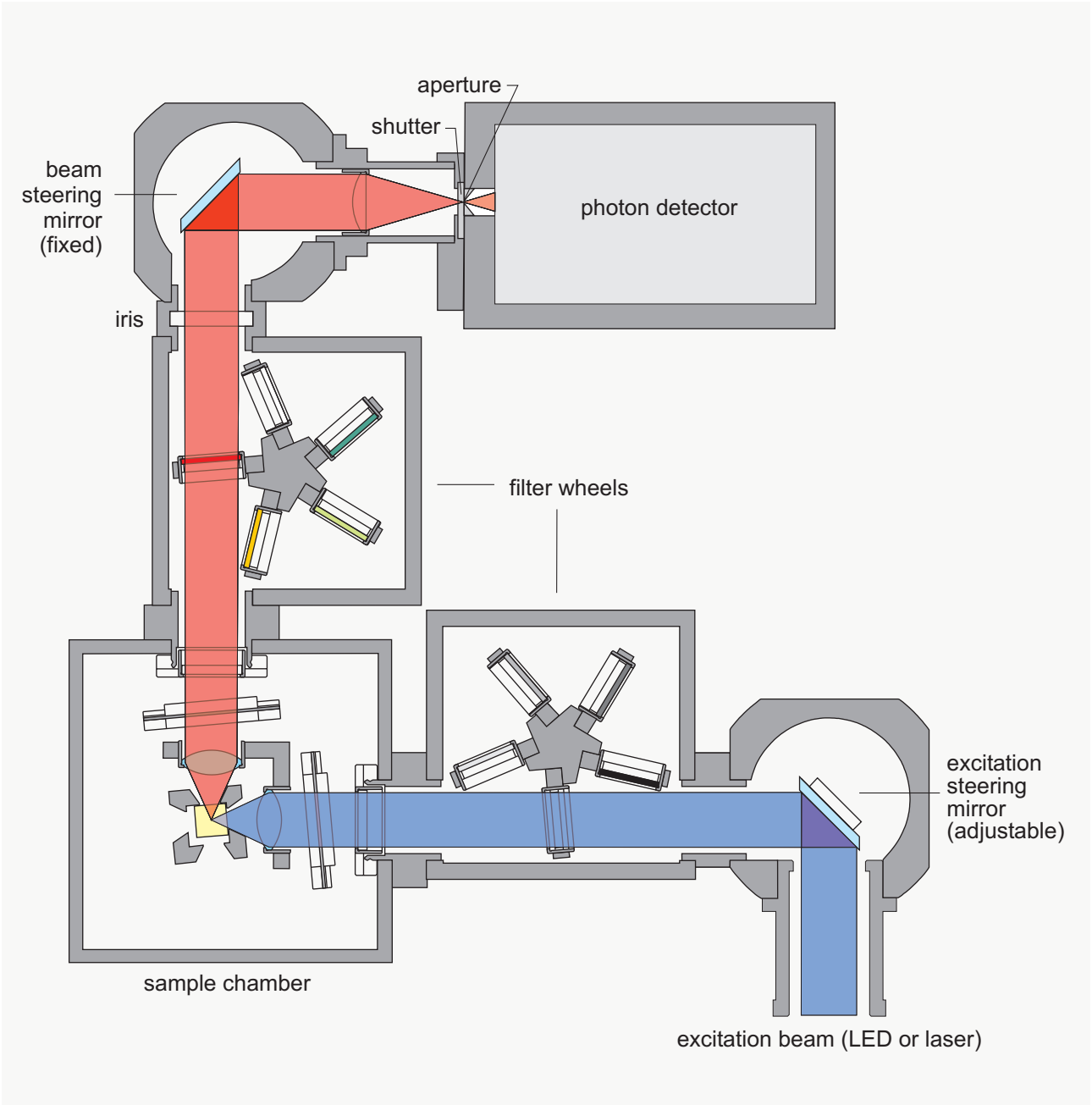


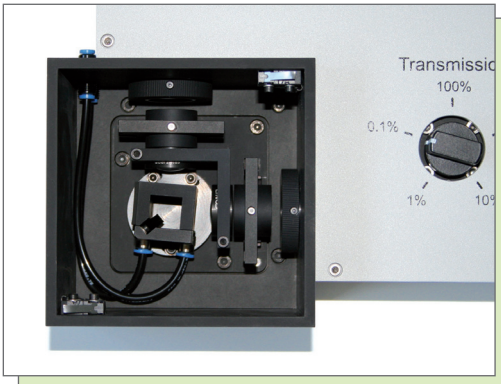
Various exponential decay models (up to fourth order) or rate constant distribution models can be fitted to the observed decay. FluoFit also supports the analysis of anisotropy measurements as well as global analysis for all methods. Running FluoFit simultaneously with the measurement software allows for parallel data analysis.

TCSPC data acquisition
The data acquisition units contain the complete timing electronics for Time-Correlated Single Photon Counting (TCSPC). More than 1 million counts per second can be processed. Consequently, TCSPC acquisition times shorter than 1 second can be utilized. All functions of the TCSPC modules are controlled by easy to use Windows™ based software. These functions include loading and saving experimental data, as well as setting and storing measurement parameters. For investigation of long lifetime samples (phosphorescence), a multiscaler with a time range up to seconds is available as an option.

Detector
The PMA detector unit, based on the Hamamatsu H5783/ H10721 series photosensor modules, is recommended for the majority of applications. The unit has a built-in high voltage power supply, signal pre-amplifier and a gold plated iron housing for optimal timing performance and maximum RF and magnetic shielding. With this detector unit, an Instrument Response Function (IRF) with less than 200 ps, in combination with a picosecond pulsed diode laser, can be achieved. Various different cathode types are available to cover the spectral range between 185 and 820 nm.

Optical Layout





Options and Accessories

Sheet polarizers
For fluorescence anisotropy measurements, 20 mm aperture sheet polarizers are supplied with the instrument. Optional UV optimized polarizers are available.

Front face sample holder
An adjustable front face sample holder can replace the standard cuvette holder. It allows front surface excitation and detection of the luminescence response with minimum scattered and reflected excitation light.

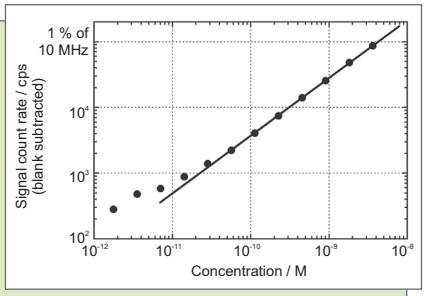
Fiber probe
The FluoTime 100 can optionally be equipped with a fiber probe for excitation and emission collection. For this purpose, the usual sample chamber is modified to hold the fiber probe. This special configuration gives even more freedom and flexibility to investigate different kinds of samples.

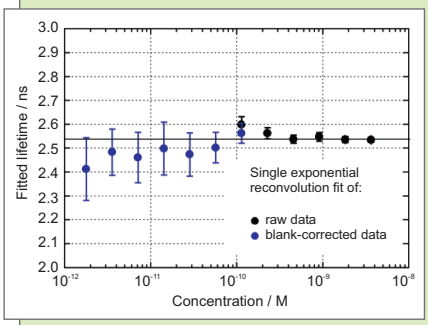
Water-cooled sample holder
The standard sample holder accommodates 1 cm (or smaller) path length cuvettes. Temperature control of the sample is possible by circulating liquid (the tubing is pre-installed). An external thermostat (optional) can be attached. The cuvette holder can be optionally equipped with a magnetic stirrer.

Filters
Under certain circumstances, the pre-installed filter set may not be sufficient for the given excitation source and fluorescent sample combination. Additional filters can be inserted into both excitation and emission arms, using the tilted filter holders in the sample chamber.

Cooled photomultiplier tube
Instead of the PMA detector assembly, the FluoTime 100 can be equipped with a Peltier-cooled Photomultiplier Tube (PMT), e.g. the H7422 detector module from Hamamatsu. Cooling the PMT considerably decreases its thermal noise, i.e. suppresses the so-called “dark counts”. The smaller dark count rate means better signal-to-noise (S/N) ratio, and the result is an ultimate sensitivity.

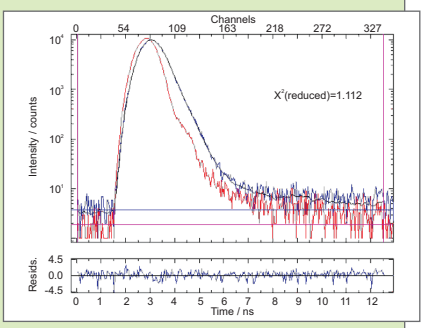
Measurement Examples

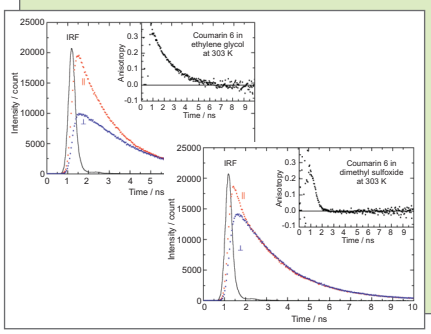




To demonstrate the sensitivity of the FluoTime 100, a set of very diluted solutions of Coumarin 6 in Ethanol was measured. The samples were excited at 10 MHz repetition rate with the LDH-P-C-405 diode laser. The average excitation intensity in the sample volume was 80 µW. Magic angle polarized fluorescence was collected through a 530 nm longpass filter. The detected intensity decreases linearly with the concentration down to 50 pM. Below this concentration, the weak, parasitic fluorescence of the glass filter must be taken into account. Performing a blank measurement with the pure solvent as a sample and subtracting this blank measurement from the TCSPC histograms allowed a single exponential lifetime analysis at picomolar concentrations.

The example on the right shows a time-resolved fluorescence measurement of Exciton LDS750 dye (10^{-7} molar in Ethanol). The sample was excited at 20 MHz repetition rate with a PLS 600 pulsed LED diode, which has a pulse width of 940 ps (FWHM). The emission was detected by the PMA 182 detector. The plot shows the measured Instrument Response Function (red), the sample decay (blue) and the fitted decay (black). The fitted fluorescence lifetime is 310 ±10 ps.





The example on the left shows a time-resolved anisotropy measurement of Coumarin 6 in ethylene glycol (EtGly) and dimethyl sulfoxide (DMSO) solvents at a temperature of 303 K. The sample was excited with the LDH-P-C-405 diode laser and the emission was detected with the PMA 182 detector. The FluoTime 100 was equipped with sheet polarizers. The figures show the recorded polarized decays, the IRF of the measurement and the calculated fluorescence anisotropy decay. Fitting the anisotropy decay to a single exponential model without reconvolution yields 1.9 ns and 270 ps rotational correlation time of Coumarin 6 in EtGly and DMSO, respectively. The results reflect the different viscosity of the solvents.

Specifications

System

Optical configuration	L-Geometry
Mode of operation	Time-Correlated Single Photon Counting (TCSPC)

Excitation sources

Light source	pulsed LEDs (PLS Series)	laser diode heads (LDH Series)
Wavelengths	245-600 nm	266-1550 nm
Pulse width	400 ps - 1 ns	60-500 ps
Repetition rate	up to 40 MHz	up to 40 MHz (optional 80 MHz)

Detectors

Type[1)]	PMT (PMA Series)	
Spectral range	185-750 nm	185-820 nm
Dark counts (at 20 °C, typ. value)	< 50 cps	< 900 cps

Data acquisition unit

Type	PicoHarp 300	TimeHarp 260	TimeHarp 260 NANO
Number of time channels/curve	up to 65536	up to 32768	up to 32768
Count depth	16 bit	32 bit	32 bit
Time resolution (bin width)	4 ps	25 ps, 2.5 ns (long range mode)	1 ns
Dead time	< 95 ns	< 25 ps, 2.5 ns (longe range mode)	< 1 ns
Full scale time range	260 ns - 33 µs	819 ns - 1.71 s 81.92 µs - 171 s (long range mode)	32.8 µs - 68.48 s

Data analysis software

Type	FluoFit (FluoFit Basic, FluoFit Professional)
Analysis possibilities	exponential decay, lifetime distribution, anisotropy, global analysis for all methods

Operation

Operating system	Windows™ 2000, XP, Vista, 7, 8

Electrical & dimensional

Power requirements	220/240 or 110/120 VAC, 50/60 Hz
Dimensions	300 × 200 × 100 mm (w × d × h)

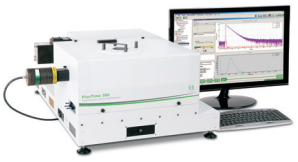
[1)] other detectors and cooling available upon request

Other PicoQuant Systems

LSM Upgrade Kit
Compact lifetime and FCS upgrade kit for Laser Scanning Microscopes

MicroTime 200
Inverse time-resolved fluorescence microscope

FluoTime 300 “EasyTau”
Automated Fluorescence Lifetime Spectrometer



FluoTime 200
High performance fluorescence lifetime system

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