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Redefining Measurement

SWISS QUANTUM

SINGLE-PHOTON SYSTEMS APPLICATION NOTE

Fluorescence Lifetime Measurement

ID100-20 Single Photon Detection Module

The advantages brought by the ID100-20 module (see Fig.1) over commercially available photomultiplier tubes (PMTs) and single photon counting modules for fluorescence lifetime measurements.

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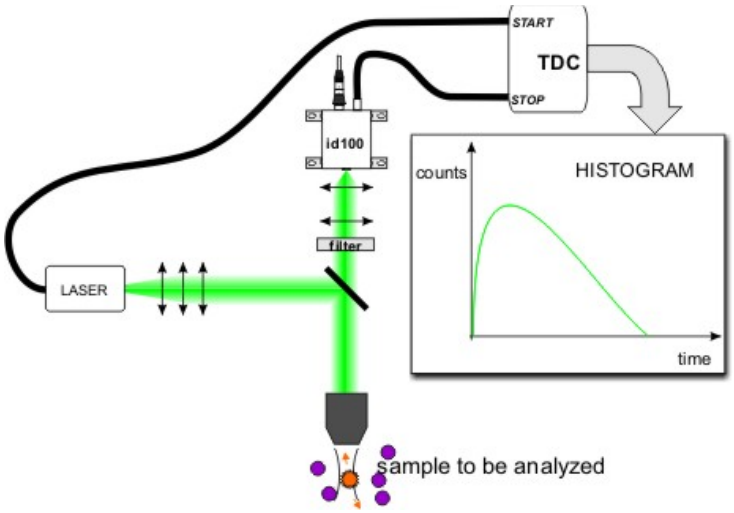
Introduction

Fluorescence lifetime measurements based on near field or confocal microscopy find applications in various fields such as biochemistry and analytical chemistry, medicine, pharmacology, photophysics and photochemistry, and environmental research. Over the past few years we have witnessed a tremendous surge of interest in single molecule detection (SMD) by Time Correlated Single Photon Counting technique (TCSPC). In single molecule detection applications, the molecules to be detected are fluorescently labelled. By repetitive laser excitations, the fluorescent molecule is cycled between its ground state and the excited state. The excited state exists for a finite time, i.e. the fluorescence lifetime during which the energy is partially dissipated until the relaxed singlet excited state is reached. Then, the fluorophore returns to its ground state by fluorescence, i.e. by emission of a photon of wavelength fluo. After many excitation pulses, the fluorescence lifetime, i.e. the average time the fluorophore spends in the excited state, is obtained, allowing the identification of the molecule.

Fig. 1: Photomicrograph of the ID Quantique's ID100-20

A typical set-up (see Fig.2) for fluorescence decay time measurement includes a pulsed laser diode, appropriate lenses for beam expansion, a microscope objective to focus the excitation beam and collect the emitted fluorescence photons, a set of appropriate lenses to focus the light on the ID100-20 photosensitive area and a bandpass filter centered on λ_{fluo}. A time-to-digital converter (TDC) or time-to-amplitude converter combined with a multichannel analyzer measures the time between the laser excitation and the fluorescent photon emission. A histogram is progressively built and the fluorescent decay time is extracted.

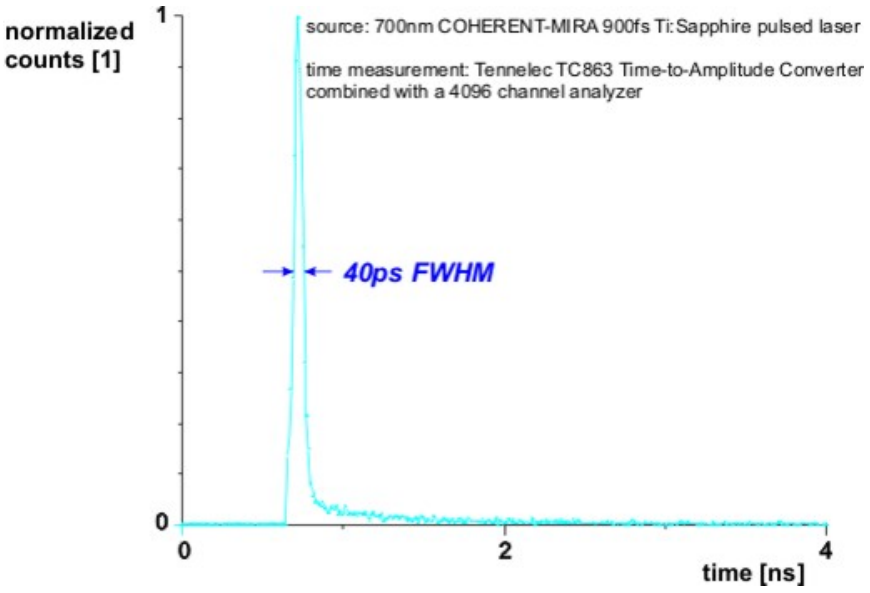
Fig. 2: Typical set-up for fluorescence lifetime measurement.



Benefits of the ID100-20

✔ A best-in-class timing resolution of 40ps FWHM (see Fig.3). This is a significant improvement compared to photomultiplier tubes and other commercially available photon counting modules. This excellent feature allows accurate measurements of extremely short fluorescence lifetimes of a few tens of picoseconds. Furthermore, multi-component fluorescence decay time experiments become possible with high resolution.

Fig. 3: Timing resolution of the ID Quantique's ID100-20.



✔ A large range of sensitivity. With a maximum in the green and a single photon sensitivity covering the entire visible range (400 to 750nm), ID Quantique's ID100-20 is suited for most fluorophores used in life sciences (see http://microscopy.bio-rad.com/fluorescence/fluorophoradata.htm). The ID100-20 photon detection probability compares favorably with standard PMTs.

✔ A dead time of 70ns, which allows a maximum excitation frequency of 14MHz. The histogram can thus be recovered in a very short time.

✔ A low rate of spurious counts including afterpulses and dark counts that usually damage the signal-to-noise ratio of your measurement. The low dark count and afterpulsing rates are obtained by limiting the active area diameter to 20mm. A carefully studied optical set-up is thus required to focus the fluorescence light onto the detector active area. ID Quantique recommends to mount the ID100-20 on a 3-axis translation stage using the available mounting brackets. A precise alignment is thus possible and only needs to be done once. Due to the limited detector area, the presence of a pinhole in front of the detector is often unnecessary. ID Quantique also offers a fiber-coupled version of the ID100-20 with a 50µm-core multimode fiber.

✔ A compact design and a high level of reliability. In addition, the ID100-20 is not damaged by ambient light and is immediately operational as no cooling is required.

Parallel Measurements

ID Quantique has introduced the ID150. This 1x10 linear array of single photon detectors is the first commercially available product allowing high throughput parallel photon counting and timing measurements (http://www.idquantique.com/photon-counting/photon-counting-modules/ID150/).

For further information on our products, please contact ID Quantique by phone: +41 (0)22 301 83 71, fax: +41 (0)22 301 83 79, or email: sales@idquantique.com. Note that ID Quantique also offers photon counting modules operating in the infrared and short-pulse laser sources. Custom developments of single photon sensors including arrays are possible.