PIE-FRET MICROSCOPY WITH FLAVIS

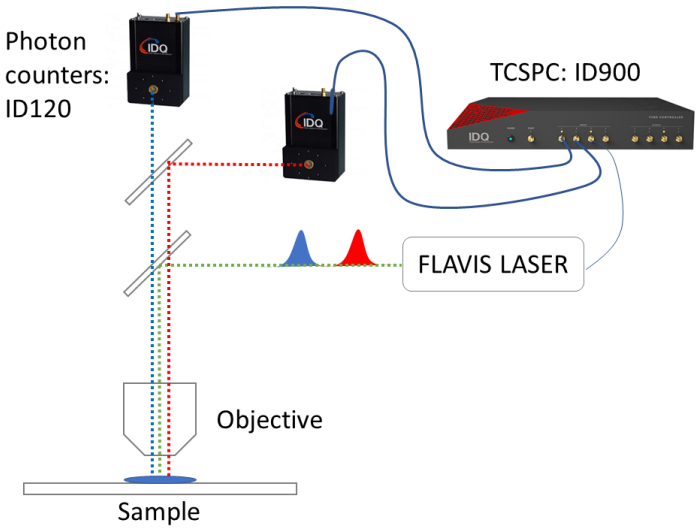
Redifining Measurement

Introduction

FRET (Förster Resonance Energy Transfer) microscopy has been a major breakthrough for dynamic observation of proteins interaction occurring at distances of a few nanometers between the molecules. The concept is to excite a “donor” fluorophore from a molecule and expect that the energy will be transfer to an “acceptor” fluorophore hold by a close neighbor molecule. This process would only occur is the donor-acceptor separation is in the order of 10 nm or less.

In addition, PIE (pulsed-interleaved excitation)-FRET allows one to access the full information of the molecules under investigation. Both acceptor and donor fluorophore are excited sequentially with 2 different wavelengths (using FLAVIS laser) resulting on usual fluorescence (or not), witnessing the presence of the acceptor and donor on the molecules. A FRET fluorescence on the acceptor molecule gives the information of the molecules distance.

Experimental scheme



We describe a potential setup to realize PID-FRET using FLAVIS Laser and ID Quantique products. The sample is getting excited by the temporally and spectrally separated pulses generated by FLAVIS. The feedback from the sample is then spectrally divided and detected with single photon counters ID120.

TCSPC (Time Correlated Single Photon Counting) is then performed between a reference signal coming from the laser and the 2 detections signals coming from the single photon counters.

The unique ability of the FLAVIS laser to generate short pulses at two different wavelength makes it especially suited for PIE-FRET applications.

Schweizerische Eidgenossenschaft
Confédération suisse
Confederazione Svizzera
Confederaziun svizra

Swiss Confederation

Innosuisse – Swiss Innovation Agency

Funded by Innosuisse

ID QUANTIQUE SA
Chemin de la Marbrerie 3
1227 Carouge/Geneva
Switzerland
T +41 22 301 83 71
F +41 22 301 89 79
info@idquantique.com
www.idquantique.com