240920

KEX-project at Scilifelab (next to Karolinska Institutet), Department of Applied Physics, KTH:

"Building a Fluorescence Correlation Spectroscopy (FCS) instrument"

FCS is a technique that analyzes concentrations, sizes, fluctuations and interactions of biomolecules, usually proteins, in solution or in living cells. As the biomolecules diffuse through a focused laser beam of ~0.4 μm diameter, they give rise to fluorescence bursts, which are analyzed by auto- or cross-correlation to give information about the concentration, sizes etc.

The task is to build an FCS instrument. If time is given, the instrument can then be used to test the boundaries of a newly developed variant called FRET-FCS (Förster Resonance Energy Transfer). The task may thus involve a little bit of research, but this is up to the students.

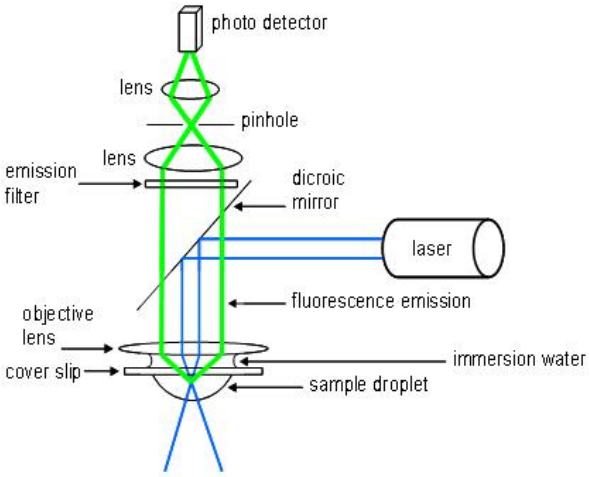

Figure 1. Principle of an FCS setup. The laser light (blue) is reflected by a dichroic mirror into a microscope objective – the objective lens in the figure – which focuses the laser light inside the sample droplet. Fluorescence light (green) is generated from dye-labeled protein molecules during the short time that they reside in the laser focus and is detected by an APD detector after passing through a pinhole in the image plane.

Building the instrument is a good way of getting acquainted with the technique. Research on FRET-FCS is needed to understand the capacity of this new FCS-variant. We know already that it has a unique ability to detect rare oligomers of the Amyloid Beta peptide in solution (Alzheimer's disease) when they are FRET-active (Wennmalm et al, *Analytical Chemistry*, 2015, Nordahl et al, *Biochimica et Biophysica Acta*, 2024). We would now like to know the detection limit of FRET-FCS, i.e. how small fractions of FRET-active oligomers that can be detected.

At the start of the project we will decide if the students want to focus on building the FCS setup, in which case they will get less assistance with that, or if they want to focus on research on FRET-FCS, in which case they will get more assistance in the building-phase.

You are welcome to call me with any questions, Stefan Wennmalm 073 712 14 39.