



Single Molecule Spectroscopy and its Applications to Nanoscience and Chemical Biology

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The course will consist of 20 h of lectures, and a final exam (2 h in class). Five different sections are identified. Section 1 will be based on standard Photochemistry books such as Modern Molecular Photochemistry of Organic Molecules. Section 2 will be based on the book Principles of fluorescence. Section 3 will be based on recent reviews on single molecule microscopy and setups. Sections 4 and 5 will be based on recent literature.

1. Introduction to Photochemistry and Photophysics (3 h)
 - a. Jablonski diagram
 - b. Singlets and triplets
 - c. Chemical vs. physical processes
 - d. Fluorescence and phosphorescence
 - e. Energy Transfer, electron transfer and singlet oxygen sensitization

2. Fluorescent probes (3 h)
 - a. Standard fluorophores
 - b. Fluorescent proteins
 - c. Emission quantum yield, absorption and brightness
 - d. Intrinsic photostability and protocols to enhance emissive properties
 - e. Fluorogenic probes, exploiting nonradiative pathways towards enhancement.

3. Microscopy setup and sample preparation in single molecule (3 h)
 - a. Microscope hardware
 - b. Excitation sources. Lasers vs. lamps, single mode vs. multimode
 - c. Wide field, Confocal and Total Internal Reflection (TIRF) microscopy

- d. Fluorescence Lifetime Imaging (FLIM)
 - e. Detectors appropriate for single molecule studies. CCDs and APDs
 - f. Sample preparation, immobilization techniques.
4. Single Molecule Super resolution microscopy (3 h)
 - a. Far field optical resolution, the gaussian profile and the centroid.
 - b. FIONA (Fluorescence Imaging with One Nanometer Accuracy)
 - c. Exploiting blinking, bleaching photoactivation and stimulated emission
 - d. Concepts behind STORM (Stochastic Optical Reconstructon Microscopy)
 - e. 2D STORM and 3D STORM
 - f. Concepts behind PALM (Photo-Activated Localization Microscopy)
 - g. Concepts behind STED (Stimulated Emission Depletion microscopy)
 - h. Concepts behind SOFI (Super-resolution Optical Fluctuation Imaging).
 5. Applications of Single Molecule to chemistry and chemical biology (6h)
 - a. Enzyme reactions
 - b. Catalysis in zeolites
 - c. Diffusion in polymer films
 - d. Au nanoparticle catalysis
 - e. Electrochemical reactions
 - f. Photocatalysis studied via single molecule

Lectures: 20 h

Personal Study: 6 h

Final Exam: 2 h

Total Course-Time: 28 h

Literature:

- Modern Molecular Photochemistry of Organic Molecules. N. Turro, University Science Books, 1991.
- Principles of Fluorescence Spectroscopy. J. R. Lakowicz, 3^a Ed., Springer, 2006.

Informations:

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