

# UNIVERSITÀ DEGLI STUDI DI MILANO-BICOCCA

# **COURSE SYLLABUS**

# **Biophotonics**

2021-1-F1701Q125

# Aims

Knowledge of the main spectroscopic techniques for the characterization of biosystems.

## Contents

- UV-Visible radiation interaction with biomolecules at the fundamental state: absorption spectroscopy.
- Fluorescence spectroscopy, spontaneous emission coefficient, Stickler-Berg expression, Stokes shift, fluorescence lifetime, quantum yield. Methods for time resolved fluorescence detection.
- Concepts of optical microscopy, imaging, resolution limit and Point Spread Function of a microscope.
- FRET (fluorescence resonant energy transfer) between two fluorophores (Forster theory) with application to microscopy.
- Fluorescence Polarization anisotropy (steady state and time-resolved), molecular form factors.
- Fluorescence fluctuations correlation techniques: FCS in solution (diffusive motions, binding kinetics, photodynamics). Temporal image correlation (TICS), space and space-temporal correlation for cellular motions detection. Flow measurements by correlation techniques.
- Super-resolution microscopy techniques:STED, STORM and PALM.
- Analysis of stochastic processes in biophysics.

## **Detailed program**

- Introduction to absorption spectroscopy UV-VIS: semiclassical model, derivation of the transition dipole strength. Protein and nucleic acid absorption, Solvent effects. Interaction among the chromophores: excitonic effect (examples) ipocromism (examples)
- Fluorescence spectroscopy: semiclassical model for the light0matter interaction: spontaneous emission coefficient, quantum yield, fluorescence lifetime. Jablonski diagrams, Stickler-Berg rule. Intrinsic fluorophores in biomolecules and tissues, probes.
- Time resolved fluorescence, methods and instrumentation.
- Notes on fluorescence microscopy, spatial resolution of an optical microscope,
- FRET, Forster theory, derivation and application in fluorescence microscopy
- Fluorescence anisotropi, strada state and time -resolved. Shape effect of biomolecules.
- Fluorescence correlation spectroscopy: principles and applications. Derivation of the expression for brownian diffusion, drift motion, chemical reaction and photodymanics.
- Image correlation: temporal correlation (TICS) and spatio-tenporal correlation(STICS): derivation an applications.
- Super-resolution techniques: STED, PALM, STORM
- Optical microscopy, an introduction
- stochastic equations for Biophysics.

#### **Prerequisites**

Knowledge of the basic concepts of quantum mechanics atomic physics achieved during the bachelor degree.

## **Teaching form**

In non-emergency time: Lectures and exercises to be done by teh students.

during covid19 times the lectures will be partially in streaming (then recorded and uploaded on the elearning site) and video-recorded. Some homeworks will be suggested.

#### **Textbook and teaching resource**

Lakowicz "Principles o Fluorescence Spectroscopy"

Parson "Modern Optical Spectroscopy"

Doi&Edwards, Polymer Dynamics,

web site for java simulations: https://phet.colorado.edu/

Scientific papers selected by the teacher

Teacher's notes

#### Semester

I semester

#### Assessment method

Oral exam. The candidate can prepare a topic at her/his choice. Other questions on fundamental topics of the course will follow. The exam will typically require 30-45 min.

The requirements to be met in order to successfully pass the exam will be clearly defined during the lessons at the beginning and end of each macro-topic.

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During the Covid-19 emergency time the exams will be organized in videocalls through the Webex platfomr and scheduled on the elearning page of the course.

# **Office hours**

always, on request.