



UNIVERSITÀ  
DEGLI STUDI DI MILANO-BICOCCA

## COURSE SYLLABUS

### Biophotonics

2122-1-F1701Q125

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#### Aims

Introduction to the main spectroscopic techniques and instruments for studying biosystems. Development of optical-spectroscopical devices for biotechnology and Medicine.

#### 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.

Techniques, optical and electronic devices and methods for time resolved fluorescence detection. Discussion of optical microscopy, imaging, resolution limit and Point Spread Function of an optical microscope. FRET (fluorescence resonant energy transfer) between two fluorophores (Forster theory) with application to optical microscopy.

Fluorescence anisotropy (steady state and time-resolved), molecular form factors. Optical and electronic devices for the measurement of the Fluorescence lifetime and correlated techniques. Fluorescence fluctuations correlation techniques and devices: FCS in solution (diffusive motions, binding kinetics, photodynamics). Temporal image correlation (TICS), spatio and spatio-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 light-matter 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 anisotropy, steady 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 photodynamics.
- Image correlation: temporal correlation (TICS) and spatio-temporal correlation (STICS): derivation and applications.
- Super-resolution techniques: STED, PALM, STORM
- Optical microscopy: confocal and non-linear excitation microscopies, principles, devices and techniques.
- stochastic equations for Biophysics.

## Prerequisites

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

## Teaching form

Lectures and exercises to be done by the students.

## Textbook and teaching resource

Lakowicz "Principles of Fluorescence Spectroscopy"

Parson "Modern Optical Spectroscopy"

Doi&Edwards, Polymer Dynamics,

Scientific papers proposed by the lecturer

OpticStudio: examples developed by the lecturer

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.

## **Office hours**

always, under request.

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