UNIVERSITÀ
DEGLI STUDI DI MILANO-BICOCCA

# SYLLABUS DEL CORSO

# Laboratory of Biophotonics I

**2526-1-F1703Q016**

## Aims

1. Knowledge and Understanding
   Students will gain a solid understanding of advanced fluorescence-based techniques (e.g., fluorescence lifetime, anisotropy, FCS, DLS) and their physical foundations, with focus on their application to the characterization of biomolecules and nanostructures.
2. Applying Knowledge and Understanding
   Students will acquire practical skills in experimental design, calibration, and quantitative analysis using time-resolved fluorescence, polarization anisotropy, and correlation spectroscopy. They will conduct measurements on protein–dye systems, nanoparticles, and complex biological media.
3. Making Judgements
   Students will develop the ability to critically assess data quality, interpret results in terms of photophysical and molecular models, and identify sources of experimental uncertainty. They will evaluate physical parameters such as binding constants, diffusion coefficients, and aggregation states.
4. Communication Skills
   Students will effectively present experimental data and scientific interpretations through technical reports and oral presentations, using appropriate terminology, graphical representation, and quantitative reasoning.
5. Learning Skills
   Students will be able to independently deepen their expertise in biophotonics and related experimental techniques, consulting primary scientific literature and adapting methodologies to novel research contexts.

## Contents

Time-resolved spectroscopic techniques applied to biosystems. Fluorescence lifetimes of typical fluorophores used in optical microscopy. Fluorescence anisotropy. Polarized and depolarized dynamic light scattering. Fluorescence correlation spectroscopy.

## Detailed program

Fluorescence lifetimes measurements of typical fluorophores used in optical microscopy. Dyes in solution and mixtures of dyes. Fluorophore-protein binding constant evaluation from lifetimes measurements. Proteins size and aggregation studies by means of fluorescence polarization anisotropy. Polarized and depolarized dynamic light scattering. Temperature and salt concentration effects on protein diffusion dynamics. Aggregation kinetics. Fluorescence correlation spectroscopy (FCS): calibration of the optical setup, experiments versus excitation power and concentration. Green Fluorescent Protein photophysics. Gold nanoparticles FCS. Molecular crowding experiments.

## Prerequisites

The topics covered in the different courses of the Bachelor Degree in Physics.

Recommended: the Biophotonics course of the Master Degree in Physics and/or the Experiments of Biophotonics course of the Bachelor Degree in Physics.

## Teaching form

Approximately eight hours of initial instructional teaching, followed by interactive laboratory teaching, with practical training activities conducted in person in the research laboratories of the Biophysics group, rooms 4054-4052-4051.

## Textbook and teaching resource

C.R.Cantor and P.R.Schimmel, “Biophysical Chemistry”, W.H. Freeman & Co, 1980;

J.R.Lackowicz, “Principles of Fluorescence Spectroscopy”, Springer, 2006;

A.Diaspro, “Confocal and two photon microscopy: foundations, applications and advances” edited by Alberto Diaspro, Wiley, 2002.

## Semester

I semester.

## Assessment method

Students are required to write a report in English describing the experiments performed and it will be the focus of the final oral examination.
Each student will also have to prepare a short presentation (10 min) on one of the experiments carried out during

the course.

The final grade will be determined by the evaluation of the report, of the knowledge of the different topics covered in the lab, of the experimental data analysis and of the student’s behavior throughout the course in the laboratory.

## Office hours

By appointment.

## Sustainable Development Goals

GOOD HEALTH AND WELL-BEING | QUALITY EDUCATION | DECENT WORK AND ECONOMIC GROWTH | INDUSTRY, INNOVATION AND INFRASTRUCTURE