

# UNIVERSITÀ DEGLI STUDI DI MILANO-BICOCCA

# SYLLABUS DEL CORSO

# Spettroscopia per le Biotecnologie

2324-3-E0201Q077

#### **Aims**

The goal of the course is to provide basic knowledge on the main spectroscopic and microscopic approaches for the characterization of the structural properties and/or the morphology of biological systems, from biomacromolecules to intact cells and tissues.

Knowledge and understanding.

After completion of the course, the student will gain knowledge of the main spectroscopic and microscopic approaches of relevance in the biotechnology field.

Applying knowledge and understanding.

After completion of the course, the student will gain knowledge of the potential applications of the presented methods in different areas, from the characterization of isolated biomolecules to that of intact cells and tissues. Making judgements.

After completion of the course, the student will be able: (i) to read and understand scientific manuscripts where results obtained by the above methods are presented; (ii) to choose suitable spectroscopic and microscopic approaches to study biological samples in consideration of their peculiar properties and of the specific aims of the investigation; (iii) to interpret the experimental data obtained using the methods discussed in the course.

Communication skills.

Use of an appropriate scientific vocabulary for the field.

Learning skills.

The knowledge on the main spectroscopic and microscopic approaches will facilitate the understanding of more advanced approaches and/or of new applications.

#### **Contents**

The course will illustrate the main spectroscopic and microscopic approaches for the study of biological systems, from biomolecules to intact cells and tissues. Applications of these methods in biotechnology, biochemistry and biomedicine will be presented. The practical aspects of these methods will be also discussed.

The course includes 10 hours of experimental work using advanced spectroscopy methods. In particular, studies of conformational properties and interactions of proteins will be performed by circular dichroism, infrared and fluorescence spectroscopies.

#### **Detailed program**

Chapter 1: Bio-Spectroscopy.

General introduction to electromagnetic waves and to the interaction between matter and radiation.

UV and visible absorption of biomolecules. Circular Dichroism.

General principles of fluorescence. Fluorescence of proteins and of the main fluorescent dyes of biotechnological interest. The effects of the environment, quenching processes, the Förster resonance energy transfer (FRET) process and fluorescence anisotropy.

Raman and infrared spectroscopy of biosystems.

Applications of these methods for the study of biomolecules and of bioprocesses in vitro and in vivo will be presented. Some examples are indicated below:

folding, stability and aggregation of proteins and peptides of biotechnological interest (biopharmaceuticals, enzymes, inclusion bodies, bio-materials, etc);

protein misfolding and amyloid aggregation (amyloid proteins involved in neurodegenerative diseases and systemic amyloidoses, fibrillogenesis in vitro and in situ and the role of the environmental factors, such as lipids and metals); characterization and monitoring of biotechnological processes in vitro and in situ, such as lipid accumulation in intact microbial cells and enzymatic transesterification for biofuel production;

biomolecular interactions, such as the study of protein-ligand and protein-nanoparticle interactions.

#### Chapter 2: Microscopy.

The design of the conventional optical microscope and of fluorescence microscope will be presented. Laser scanning confocal fluorescence microscopy and its spatial resolution will be examined with the applications on single cells. Advanced super-resolution microscopies will be also discussed, with particular emphasis on STED microscopy.

The Atomic Force Microscopy (AFM), Electron Microscopies (TEM, SEM and Cryo-EM) and microspectroscopic approaches will be also presented.

Their applications for the study of biomolecular complexes, cells, intracellular structures and sensors, and tissues will be discussed.

#### Chapter 3: Laboratory activities.

Conformational changes and aggregation of proteins studied by circular dichroism and infrared spectroscopies. Biomolecular interactions studied by circular dichroism and fluorescence spectroscopies.

The experimental activity involves sample preparation, spectroscopic data collection and subsequent analysis.

# **Prerequisites**

Background: basic knowledge of Biochemistry.

Specific prerequisites: none.

General prerequisites: Students can take the exams of the third year after having passed all the exams of the first year of the course.

#### **Teaching form**

Classroom lessons supported by PowerPoint presentations, and interactive discussions with students on the interpretation of experimental data.

Teaching language: italian.

# Textbook and teaching resource

Learning material (slides of the lessons, suggested websites and recommended scientific publications) is available at the e-learning web page of the course.

Recommended textbook:

Dagmar Klostermeier, Markus G. Rudolph, Biophysical Chemistry CRC Press; December 19, 2017

#### Semester

First semester

#### Assessment method

Oral examination.

The examination consists of three parts:

- (50% of the final grade) Interpretation of experimental data;
- (30 % of the final grade) assessment of the basic knowledge of at least two spectroscopic methods among those presented;
- (20% of the final grade) assessment of the basic knowledge of one of the microscopic methods among those presemted.

#### Office hours

Contact: on demand, upon request by mail to lecturer.

# **Sustainable Development Goals**

GOOD HEALTH AND WELL-BEING | QUALITY EDUCATION