



**UNIVERSITÀ
DEGLI STUDI DI MILANO-BICOCCA**

SYLLABUS DEL CORSO

Introduzione alle Tecniche di Laboratorio

2526-2-E1301Q079

Aims

The primary objective of this course is to provide students with a basic understanding of the most common biochemical and recombinant DNA techniques. It also aims to illustrate the complementarity between cloning strategies, the production, purification, and characterization (structural and functional, with particular emphasis on enzymatic activity) of recombinant proteins.

Knowledge and understanding - At the end of the course, the student will be familiar with the theoretical principles of the main biochemical laboratory techniques and basic recombinant DNA manipulation techniques.

Applying knowledge and understanding - At the end of the course, the student will be able to apply the acquired knowledge in choosing an experimental approach for DNA cloning, protein production, purification, and characterization. This knowledge will also be applied in subsequent courses, particularly in the Integrated Biology Laboratory (LIB) course.

Making judgments - At the end of the course, the student will be able to recognize and understand the main steps of a plasmid DNA cloning protocol, recombinant protein production, purification, and structural characterization of proteins.

Communication skills - At the end of the course, the student will have acquired an adequate technical-scientific vocabulary and will be able to properly explain the topics covered in the course.

Learning skills - At the end of the course, students are expected to be able to understand and apply the learned methodologies in different contexts (e.g., in scientific articles, experimental reports and protocols).

Contents

1. Recombinant DNA methods for cloning and producing recombinant proteins.

2. Preparative techniques for protein extraction and enrichment.
3. Electrophoretic and immunochemical techniques.
4. Techniques for protein and enzymatic activity assays.
5. Preparative techniques for protein purification.
6. Selected spectroscopic techniques for protein conformational analysis.

Detailed program

1. Recombinant DNA methods for cloning and producing recombinant proteins. Recombinant protein production strategy; Gene cloning with restriction and ligation enzymes; Choice of vectors and cloning hosts; DNA amplification (polymerase chain reaction); Plasmid DNA extraction methods; DNA electrophoresis; Capillary electrophoresis; DNA sequencing with Sanger's method.

2. Preparative techniques for protein extraction and enrichment. Centrifugation and ultrafiltration techniques; Ammonium sulfate precipitation and fractionation; Cell lysis and differential centrifugation techniques.

3. Electrophoretic and immunochemical techniques. Electrophoresis under native and denaturing conditions (SDS-PAGE); Western blotting; Introduction to immunochemical techniques; Immunoprecipitation, ELISA.

4. Techniques for protein and enzymatic activity assays. Protein concentration assay; Enzymatic activity assays; Purification table.

5. Preparative techniques for protein purification. Introduction to chromatographic techniques; Size exclusion chromatography; Ion exchange chromatography; Hydrophobic interaction and reverse phase chromatography; Affinity chromatography; HPLC and FPLC; Evaluating purification progress.

6. Biophysical techniques for protein conformational analysis. Introduction to spectrophotometry and UV-Vis absorption; Circular dichroism spectroscopy; Spectrofluorimetry; Fluorescence resonance energy transfer (FRET) techniques.

Prerequisites

Basic knowledge of physics, general, and organic chemistry is required. The physico-chemical principles and essential biochemical concepts for understanding the different methodologies will be briefly explained or reviewed at the beginning of the lessons.

Teaching form

The course comprises 24 lessons (6 ECTS) delivered in a blended format. These lessons will be both 'traditional' (conventional, in-person delivery) and 'interactive,' fostering active student participation in the analysis and interpretation of experimental methods. Learning materials will include protocol books, scientific methodology journals, and research articles.

Each lesson will be supported by PowerPoint presentations, videos, and analysis of experimental methods. Based on the requests of attending students, new study topics may be introduced.

Depending on the number and requests of attending students, group work aimed at analyzing experimental methods and their complementarity may be carried out.

Textbook and teaching resource

Textbooks:

- K. Wilson & J. Walker (2010). Principles and Techniques of Biochemistry and Molecular Biology.
- M. C. Bonaccorsi di Patti, R. Contestabile, M. L. Di Salvo "Metodologie Biochimiche" Casa Editrice Ambrosiana, 2012

****Teaching materials ****available on the course Moodle page (<http://elearning.unimib.it/>):

- Lecture slides
- Exam questions and exercises
- Scientific articles selected by the teacher.

Semester

Second semester

Assessment method**Written + oral examination.**

The exam consists of a written test (60 minutes) and an oral examination (approx. 20 minutes).

The written test includes multiple-choice questions and exercises, with a maximum score of 15 points. To be admitted to the oral examination, a minimum score of 10 points is required.

The oral examination will consist of two to three questions covering the entire program, with a maximum score of 15 points.

The final grade is the sum of the scores obtained in the written and oral tests. The passing threshold for the exam is set at 18 points.

Both assessments aim to evaluate theoretical knowledge, the ability to interpret experimental data, and to establish relationships between different techniques and experimental approaches. Communication skills and the use of scientifically and technically appropriate language will also be assessed.

There are no in-progress tests.

Office hours

By appointment, via email to stefania.brocca@unimib.it

Sustainable Development Goals

QUALITY EDUCATION
