

UNIVERSITÀ DEGLI STUDI DI MILANO-BICOCCA

SYLLABUS DEL CORSO

Metodologie Biochimiche e Tecnologie Biomolecolari

2122-2-E0201Q050

Aims

The course aims at providing the principles, implementation and applications of the main biochemical methods for the study of proteins and the manipulation and analysis of nucleic acids. Particular attention is given to the discussion of the chemical-physical processes on which the treated techniques are based and to the description of the main experimental strategies in which they are inserted.

Knowledge and understanding.

Students will gain knowledge of the fundamental principles at the basis of the main biochemical and biomolecular techniques for protein studies and manipulation of nucleic acids, as well as their implementation and application.

Applying knowledge and understanding.

Students will be able to apply the acquired knowledge to the subsequent laboratory activities and courses, as well as to understand and interpret the scientific papers required for the preparation of their graduation thesis.

Making judgements.

Students will be able to process the acquired knowledge, in order to interpret and critically discuss results from the literature and their own work, obtained by the described methods.

Communication skills.

Students will be able to use an appropriate scientific language in the description of the acquired methods and in the discussion of the experimental results.

Learning skills.

Students will have the skills in reading and understanding to face autonomously the subsequent studies that require knowledge in biochemical and biomolecular methods and will be able to apply the acquired study method

Contents

Module 1 - (Biochemistry methods in Biochemistry). PREPARATION OF PROTEIN RAW EXTRACT.

PROTEIN PURIFICATION AND ANALYSIS.

BIOPHYSICAL CHARACTERIZATION OF PROTEINS BY SPECTROSCOPIC AND SPECTROMETRIC METHODS.

Module 2- (Biomolecular techniques) RECOMBINANT DNA TECHNIQUES. CLONING AND SCREENING.

Detailed program

Module 1 - (Biochemistry methods in Biochemistry).

PREPARATION OF PROTEIN RAW EXTRACT. Salts, buffers, detergents, denaturating agents, methods for cell lysis, protein precipitation in ammonium sulfate, dialysis.

PROTEIN PURIFICATION AND ANALYSIS. Centrifugation, chromatography, electrophoresis, Western blot, immunoprecipitation.

BIOPHYSICAL CHARACTERIZATION OF PROTEINS BY SPECTROSCOPIC AND SPECTROMETRIC METHODS. UV-Vis absorbance, circular dichroism, fluorescence, mass spectrometry, surface plasmon resonance.

Module 2- (Biomolecular techniques)

RECOMBINANT DNA TECHNIQUES. Nucleases (DNase, RNase, S1 nuclease and restriction enzymes), methylases, DNA ligase, phosphatases, T4 polynucleotide kinase, DNA polymerases and RNA polymerases. Restriction maps. Agarose gel electrophoresis. Polymerase chain reaction (PCR): principles and applications. DNA probes (5' and 3' end labelling, nick translation, random priming and PCR). RNA probes. Southern and Northern analyses. DNA sequencing: basic concepts, Sanger sequencing, automatic sequencing and pyrosequencing. CLONING AND SCREENING. Escherichia coli as a host organism. Selection markers. Cloning vectors. Genomic libraries, cDNA and expression libraries. Screening.

Prerequisites

Background. Basics of Physics, General Chemistry, Bichemistry and Molecular Biology.

Specific prerequisites: None

General prerequisites: Students can take the exams of the second year after passing the examinations of Introductory Biology, General and Inorganic Chemistry, Mathematics, and Foreign Language.

Teaching form

Classroom lectures carried out with the support of slides and exercises on the blackboard.

Teaching language: italian.

Textbook and teaching resource

Learning material is available at the e-learning platform of the course.

Recommended textbooks:

- Bonaccorsi Di Patti M.C., Contestabile R. & Di Salvo M.L. "Metodologie biochimiche". Ambrosiana
- Dale J.W. & von Schantz M. "Dai Geni ai Genomi". EdiSEs
- Brown T.A. "Biotecnologie Molecolari". Zanichelli

Semester

Second semester

Assessment method

Written examination (2 h).

Two sets of open questions on the content of both course modules. The final grade will be defined as the average of the two sets, provided they both have a sufficient mark (18/30).

Office hours

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

Grandori. Contact hours: Monday 4-5 p.m. or upon request by mail.