UNIVERSITÀ
DEGLI STUDI DI MILANO-BICOCCA

COURSE SYLLABUS

Biochemistry Practical Course

2425-2-E0201Q052-E0201Q063M

Aims

The course aims to provide students with the theoretical and practical knowledge to apply experimental methodologies of biochemistry and cellular biochemistry. In particular the students hey will perform different types of experiments that will allow them to acquire the ability to work in sterility with different mammalian cell models and to analyze their behavior in different experimental conditions. The experimental experience will be accompanied by basic notions of elaboration and analysis of the experimental results obtained during the practical course. Knowledge and understanding. To consolidate and deepen basic knowledge of theoretical, technical and methodological issues already presented by the course of biochemistry. Applying knowledge and understanding. To be able to correctly interpret the experimental protocols of biochemistry and cellular biochemistry, recognize their salient aspects, collect and process experimental data. Making judgments. To develop a critical vision of the experimental design and of the results achieved. To be able to recognize the context for appropriate application of the experimental procedures methods learned during the course. Learning skills. To be able to correctly interpret experimental protocols in contexts different from those used during the practical laboratory experience. Communication skills. To be able to elaborate experimental data and describe the results in an appropriate language and with the correct technical terms.

Contents

1. Manipulation and propagation of mammalian cells in vitro;
2. Aseptic techniques for the protection of the operator, the cell culture and the environment;
3. Cell growth curves and in vitro doubling time;
4. Use of a technique that allows the introduction of nucleic acids into mammalian cells, using a chemical method, in a process called "transfection";
5. Protein quantization by spectrophotometric assay;
6. Analysis of enzymatic activity by spectrophotometric assay;
7. Protein localization analysis by fluorescence microscopy;
8. Protein expression analysis by western blot;

9. Protein expression by fluorometric analysis;
10. Analysis of cell viability by spectrophotometric assay;
11. Analysis and critical discussion of the results obtained from the use of the above techniques.

Detailed program

1. In vitro manipulation and propagation of mouse and human cells, in particular will be used NIH3T3 cells either immortalized or transformed with the K.Ras oncogene;
2. Aseptic techniques for the protection of the operator, the product (cells) and the environment, in particular the student will be introduced to the use of a sterile biological hood, the use of sterile material and the necessary behaviors to preserve such sterility;
3. Cell growth curves and in vitro doubling time, in particular will be used NIH3T3 cells either immortalized or transformed with the K.Ras oncogene grown in different growth conditions;
4. Use of a technique that allows the introduction of nucleic acids into mammalian cells, using a chemical method, in a process called "transfection", in particular the technique of Calcium-Phosphate will be used;
5. Protein quantization by spectrophotometric assay, in particular the student will evaluate the protein concentration of a cell extract using the Bradford method;
6. Enzymatic activity analysis using a spectrophotometric assay, in particular the student will analyze the enzymatic activity of the beta-galactosidase protein;
7. Analysis of protein expression by fluorescence microscopy, in particular the student will analyze the expression of the Green Fluorescent Protein following its expression by transfection in mammalian cells;
8. Protein expression analysis by western blot technique, in particular the student will perform the complete procedure used to analyze the expression of the Green Fluorescent Protein following its expression by transfection in mammalian cells;
9. Cell vitality analysis by MTT assay
10. Critical analysis of the results obtained through the use of the aforementioned techniques.

Prerequisites

Background: participation in the Biochemistry course.
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

Each learning unit is addressed to a group of 35-40 students, through practical lessons by interactive teaching which are carried out in a dedicated teaching laboratory. At the beginning of each lesson, theory, aims and experimental design will be exposed. At the end of each experimental part , an overall discussion of collected results will take place in the same laboratory. For further details, please, refer to lesson calendar on the website of Biotechnology course. Attendance at the laboratory is mandatory.
Teaching language: italian.

Textbook and teaching resource

Learning material (slides of introductory lessons, handout, experimental data) is available at the e-learning platform of LTA-Biochemistry module.

Semester

Second semester

Assessment method

Test with closed answers and open questions. The exam is carried out online in presence. In fact, students will access the exam through the University's Computerized Exam Platform (Moodle). The exam lasts 1 hour and 30 minutes. The test consists of a closed-ended test (20 multiple choice questions) and 2 open questions that require an articulated and detailed answer (short essay). The multiple choice and open questions are intended to verify the student's preparation for both the theoretical and practical parts both provided during the course. However, both notions are well described in the material already in the student's possession. This information will be repeated and explored in depth during discussions in the teaching laboratory.

Office hours

Contact: on demand, upon request by mail to course’s professor.

Sustainable Development Goals

QUALITY EDUCATION