COURSE DATA

DATA SUBJECT

Code: 33141
Name: Cellular analysis techniques
Cycle: Undergraduate Studies
ECTS Credits: 4.5
Academic year: 2026-27

STUDY (S)

Degree	Center	Acad. year	Period
1109 - Degree in Biochemistry and Biomedical Sciences	Facultat de Ciències Biològiques	3	Second quarter

SUBJECT-MATTER

Degree	Subject-matter	Character
1109 - Degree in Biochemistry and Biomedical Sciences	Métodos instrumentales	COMPULSORY

COORDINATION

RODRIGUEZ FERRON SACRAMENTO

SUMMARY

"Methods in Cell Biology" is a compulsory subject taught in the second semester of the third year of the degree in Biochemistry and Biomedical Sciences and corresponds to 4.5 ECTS. This subject is included in the block "Methods in Molecular Biosciences" and has a mixed theoretical-experimental nature. *Methods in Cell Biology* comprise the set of techniques and approaches for the structural-functional analysis of biological material at the microscopic level. Four aspects will be covered: instrumentation, techniques for the preparation of biological material for microscopic observation, techniques developed to obtain information on biological processes (in situ detection) and techniques for manipulating cells and tissues. In the theoretical part, the student will obtain an updated view of the observation techniques of different biological samples at the various levels of resolution offered by the current modern microscopes. In the practical part, students will get hands-on experience in the preparation of samples for their analysis at the microscope level, in the use and understanding of the equipment and in techniques for the in situ detection of molecules and processes at the cellular level.

PREVIOUS KNOWLEDGE

RELATIONSHIP TO OTHER SUBJECTS OF THE SAME DEGREE

There are no specified enrollment restrictions with other subjects of the curriculum.

OTHER REQUIREMENTS

COMPETENCES / LEARNING OUTCOMES

1101 -

Capacidad para diseñar experimentos y aproximaciones multidisciplinares para la resolución de problemas concretos.

Capacidad para presentar, discutir y extraer conclusiones de los resultados de los experimentos científicos.

Capacidad para trabajar correctamente en los laboratorios de bioquímica, genética, biología molecular y celular incluyendo seguridad, manipulación, eliminación de residuos y registro anotado de actividades.

Capacidad para utilizar la instrumentación básica en experimentación molecular y celular.

Tener una visión integrada de las técnicas y métodos utilizados en biociencias moleculares y biomedicina.

DESCRIPTION OF CONTENTS

1. INTRODUCTION TO METHODS IN CELL BIOLOGY

Introduction to methodology and samples used in cell biology. Types of samples and their handling. Cell cultures. Preparation of living samples. Imprints and smears. Subcellular fractionation. Introduction to histological technique.

2. LIGHT MICROSCOPY

Conventional light microscopy. The light microscope: theory of image formation. Elements of the microscope. Resolution. Optical aberrations and their correction: types of objectives. Dark field. Light interference. Phase contrast. Interferential phase contrast. Fluorescence. Fluorescence microscope. Confocal scanning microscopy. Microcaptura optical tweezers and laser.

3. HISTOLOGICAL TECHNIQUE

Fixing: Background and fixing utilities. Types of fixation. Chemical binding agents. Immersion and perfusion. Inclusion: A Foundation for inclusion. Means of inclusion. Embedding in paraffin. Freezing. Embedding. Microtomy: Getting sections. Microtomy of paraffin. Freezing microtome. Vibratome. Handling and mounting sections on slides. Staining of biological samples. Dyes. Conventional stains. Staining procedures and sample preparation.

4. CYTO/HISTOCHEMICAL TECHNIQUES

Classical histochemical techniques: Detection histochemistry of lipids, carbohydrates, nucleic acids and metals. Histoenzimology. Histoenzimology Utilities. Histochemical detection of enzyme reporters. Histological technique. Controls. Techniques based on fluorescence cell biology: Marking with lectins and other natural ligands. Detecting molecules by fluorophores. Detection of organelles by fluorophores. Introduction of fluorescent compounds into cells. Fluorescent Detection of receptors, cytoskeletal molecules, second messengers.

5. IMMUNOCYTO/HISTOCHEMICAL TECHNIQUES

Immunocytochemistry: polyclonal and monoclonal antibodies. Antibody preparation. Enzymatic systems and fluorescent detection. Direct methods, indirect and three layers. Histological technique. Controls.

6. IN SITU DETECTION OF CELLULAR PROCESSES

Endocytosis, intracellular trafficking and interaction of proteins: Techniques of analysis of the processes of endocytosis. Introduction of proteins in cells. Techniques to assess movement and interaction of proteins. FRET, FRAP. Assessment analysis of the polarity and adhesion. Aggregation analysis. Analysis of transmigration. Analysis of polarity. Techniques for assessing cellular proliferation. Nucleotide incorporation technique. Immunocytochemical markers of cell cycle. Measures the amount of DNA. Analysis of mitosis. Determination techniques of cell degeneration. Apoptosis and necrosis. Techniques for determination of cytotoxicity and viability. TUNEL technique. Annexin technique. Immunocytochemical markers of apoptosis. Autoradiographic techniques: radioactive isotopes. Pulse-capture experiments. Autoradiographic techniques. Radioligand receptor and determination. Radioactive techniques for cell proliferation.

7. TECHNIQUES FOR NUCLEIC ACID HYBRIDIZATION

Situ hybridization techniques: Hybridization of nucleic acids. Probes. Hybridization conditions. Detection systems. Isotopic and non isotopic techniques. FISH.

8. ELECTRON MICROSCOPY

The electron microscope: transmission and scanning EM. Elements. Resolution. Sample preparation. Ultrastructure: Fixation. Resin embedding. Ultramicrotomy. Contrast with heavy metals. Critical point. Special techniques: Immuno-histochemistry for EM. Cryo-ultramicrotomy. Freeze-fracture.

9. Program of practical sessions

1. Histological technique: fixation, embedding and microtomy. Preparation of various samples for microscopic observation. Inclusion and freezing. Paraffin microtomy, cryomicrotomy and vibratome sectioning. Handling of sections.
2. Color and fluorescence. Routine histological staining. Staining with fluorochromes.
3. Histochemistry. Techniques for the in situ detection of various molecules. Histoenzimology. Analysis of the expression of reporter genes.
4. Immunocytochemistry. Detection of antigens. Immunocytochemical methods and strategies.
5. In situ detection of cellular processes. Determination of proliferation and degeneration processes.
6. Electron microscopy. Sample preparation. Use of instruments.

WORKLOAD

PRESENCIAL ACTIVITIES

Activity	Hours
Theory	21,00
Laboratory	24,00
Total hours	**45,00**

NON PRESENCIAL ACTIVITIES

Activity	Hours
Attendance at other activities	0,00
Individual or group project	7,50
Independent study and work	30,00

Preparation of lessons	30,00
Preparation for assessment activities	0,00
Resolution of case studies	0,00
Total hours	**67,50**

TEACHING METHODOLOGY

The development of the subject is divided into:

Lectures. Presentation and discussion of previously announced selected topics. Teaching and bibliographic resources will be available to students in multimedia. The teacher will present the basics of the subject, devoting more time to highly complex issues, and will guide the students in the integration of the contents with related issues of other subjects.

Laboratory practical classes. A program of laboratory experiences will connect theoretical classes with research approaches in the field. Attendance is compulsory.

Tutorials. There will be two one-hour tutorials to discuss the development and results of the laboratory practical classes and to provide an integrated view of the methologies. During the tutorials, the progress of the course will be discussed. One of the tutorial will be also used to resolve problems related to the topics studied during the classes and the second tutorial will be a research talk.

EVALUATION

The assessment of the **theoretical course content** and **laboratory practices** will be carried out through **a single integrated exam**, which may include questions in various formats (multiple choice, open-ended, or experimental problem-solving). This exam is designed to evaluate the knowledge and understanding acquired by the student, as well as their ability to use the specific scientific language related to the subject. The grade obtained in this exam will account for **70% of the final mark**.

To prepare the **practical activity report**, the student will carry out **an exercise integrating the results** obtained during the laboratory sessions. The evaluation of the quality of this exercise, together with the monitoring of the students during class sessions, will help the instructor assess their learning progress and will represent **30% of the final mark**.

To pass the course, it is mandatory to attend practical classes, submit the written practical exercise, and obtain a minimum score of 5 out of 10 on the exam. Completing online tests may increase the final grade by up to 5%, with the aim of encouraging regular review of each topic.

REFERENCES

Most Histology and Cell Biology textbooks include a methodology chapter which, although very basic, can provide students with an initial overview of sample preparation and observation techniques.

There are also websites that offer very interesting information on microscopy and sample preparation techniques (images, protocols, etc.). These are just a few examples:

Microscopy:

http://131.229.114.77/Histology

http://www.cellsalive.com/enhance0.htm

http://micro.magnet.fsu/primer/index.html

Electron Microscopy:

http://www.bris.ac.uk/Depts/PathAndMicro/CPL/emtechs.htm

Tissue Staining:

http://members.pgonline.com/~bryand/

http://www.medinfo.ufl.edu/dental/denhisto/stains.html#AA14

http://www.bris.ac.uk/Depts/PathAndMicro/CPL/histmeth.htm

To delve deeper into each section of the program, students may consult the following specialized books and publications.

Location Key: CI: Science Library, M: Medicine Library, DEP: Department of Cell Biology

- Bancroft, J.D. and Hand, N.M. (1987) Enzyme histochemistry. Oxford University Press, Oxford (CI).
- Bozzola J.J. y Russell L.D. (1992) Electron microscopy: principles and techniques for biologists. Jones and Bartlett (eds.), Boston. (CI).
- Burck, H.C. (1969). Técnica histológica. Ed. Paz Montalbo. Madrid. (M) - Cuello (1985). Immunohistochemistry. John Wiley & Sons, New York. (DEP).
- Durfort M. et al (1991) Técnicas de inmunocitoquímica en microscopía electrónica. Publicaciones de la Universidad de Barcelona. (CI).
- Durfort, M. et al. (1990). La fixació (cinta de video). Publicaciones de la Universidad de Barcelona. (CI).
- Durfort, M. et al. (1990). La tinció (cinta de video). Publicaciones de la Universidad de Barcelona. (CI).

- Durfort, M. et al. (1990). La microtomia (cinta de video). Publicaciones de la Universidad de Barcelona. (CI).
- García del Moral, R. (1993) Laboratorio de Anatomía Patológica. Interamericana. Madrid. (DEP).
- Lacey, A.J. (1989) Light microscopy in biology: a practical approach. IRL Press, Oxford. (CI) - Locquin, M. (1985) Manual de microscopía. Labor, Barcelona. (CI)
- McManus, J.F.A. and Mowry, R.W. (1968) Técnica histológica. Ed. Atika, Madrid. (CI) - Mercer, E. H., Birbeck, M.S.C. (1974). Manual de microscopía electrónica para biólogos. Ed Blume, Madrid. (CI).
- Montuega Badía, L (2014) Técnicas en histología y biología celular. 2ª ed. Elsevier España (DEP).
- Peinado M.A. y cols. (1996) Avances en inmunocitoquímica y técnicas relacionadas. Servicio de Publicaciones de la Universidad de Jaén. (CI)
- Renau, J. y Megías, L. (1998) Manual de Técnicas de Microscopía Electrónica (M.E.T.). Aplicaciones Biológicas. Ed. Universidad de Granada. (DEP)
- Sauret, M. (1984) Microscòpia. Publicacions i Edicions de la Universitat de Barcelona. (DEP)
- Sampedro, A. et al (1995) Técnicas de fluorescencia en microscopía y citometría. Servicio de Publicaciones de la Universidad de Oviedo. (M)