66026 - Cellular separation. Cellular viability study

Información del Plan Docente

Academic Year	2017/18
Faculty / School	100 - Facultad de Ciencias
Degree	537 - Master's in Molecular and Cellular Biology
ECTS	6.0
Year	1
Semester	Indeterminate
Subject Type	Optional
Module	---

1.General information

1.1.Introduction

1.2.Recommendations to take this course

1.3.Context and importance of this course in the degree

1.4.Activities and key dates

2.Learning goals

2.1.Learning goals

2.2.Importance of learning goals

3.Aims of the course and competences

3.1.Aims of the course

3.2.Competences

4.Assessment (1st and 2nd call)

4.1.Assessment tasks (description of tasks, marking system and assessment criteria)

5.Methodology, learning tasks, syllabus and resources

5.1.Methodological overview

5.2.Learning tasks

5.3.Syllabus

The course will address the following topics:

66026 - Cellular separation. Cellular viability study

1. Lectures

1. Introduction and general comments (the bibliography needed for preparing the seminars has already been assigned to each student previously)
2. Assessment of cell functionality
3. Cell separation by chemotaxis, *swim-up* and filtration techniques
4. Cell separation by centrifugation methods
5. Flow cytometry
6. Electrophoresis of cells
7. Cell affinity separation
8. Cell partition in aqueous two-phase systems

2. Practice sessions

1. **Separation of motile cells:** Swim-up **versus Sucrose washing.**

Comparison of the efficiency of both methods following:

- Cell counting
- Motility evaluation by CASA
- Cell viability assessment by fluorescence microscopy
- Cell viability assessment by flow cytometry

2. **Cold-shock effect on cell functionality**

a) Analysis of the initial sample determining:

- Viability (CFDA/PI staining) by fluorescence microscopy and flow cytometry
- Reactive oxygen species (ROS) by flow cytometry
- Capacitation state (spermatozoa) by CTC staining and fluorescence microscopy

b) Analysis of the cold-shocked sample determining:

- Viability post- *cold-shock* (CFDA/PI staining) by fluorescence microscopy and flow cytometry
- Reactive oxygen species (ROS) post- *cold-shock* by flow cytometry
- Capacitation state post- *cold-shock* (spermatozoa) by CTC staining and fluorescence microscopy

3. **Identification of cell subpopulations by detection of specific proteins through indirect immunofluorescence**

a) Sample preparation

- Fixation and drying
- Washes
- Blocking and incubation overnight with the primary antibody

b) Analysis

- Washes
- Incubation with the secondary antibody
- Mounting and observation

4. **Cell separation by partition chromatography in aqueous two phase systems**

- Determination of the partition ratio
- Assessment of total and viable cell recovering

3. Seminars

66026 - Cellular separation. Cellular viability study

Each student must explain the background, objectives, main methods, results and discussion of the assigned article. A personal and critical explanation within the framework of the course is required. A written summary has also to be presented.

5.4.Course planning and calendar

Lectures and practice sessions will usually take place during the second forthnight of May.

Further information concerning the timetable, classroom, assessment dates and other details regarding this course, will be provided on the first day of class or please refer to the virtual platform Moodle https://moodle2.unizar.es/add/,

5.5.Bibliography and recommended resources