

## 66026 - Cellular separation. Cellular viability study

### Información del Plan Docente

Academic Year	2016/17
Academic center	100 - Facultad de Ciencias
Degree	537 - Master's in Molecular and Cellular Biology
ECTS	6.0
Course	1
Period	Indeterminate
Subject Type	Optional
Module	---

### 1. Basic info

#### 1.1. Recommendations to take this course

#### 1.2. Activities and key dates for the course

### 2. Initiation

#### 2.1. Learning outcomes that define the subject

#### 2.2. Introduction

### 3. Context and competences

#### 3.1. Goals

#### 3.2. Context and meaning of the subject in the degree

#### 3.3. Competences

#### 3.4. Importance of learning outcomes

### 4. Evaluation

### 5. Activities and resources

#### 5.1. General methodological presentation

#### 5.2. Learning activities

#### 5.3. Program

### LECTURES

1. Introduction and general comments (the bibliography needed for preparing the seminars has already been assigned to each student previously)

## 66026 - Cellular separation. Cellular viability study

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

### PRACTICAL 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

### SEMINAR SESSIONS

## 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.Planning and scheduling

The theoretical and practical sessions will usually take place during the last weeks of May or first weeks of June. The venue, timetables, etc will be published through the University Moodle platform at: <https://moodle2.unizar.es/add/>.

### 5.5.Bibliography and recommended resources