

DEGREE CURRICULUM GENETIC ENGINEERING

Coordination: GARI MARSOL, ELOI

Academic year 2019-20

Subject's general information

Subject name	GENETIC ENGINEERING							
Code	101611							
Semester	2nd Q(SEMESTER) CONTINUED EVALUATION							
Typology	Degree Course Character Modality							
	Bachelor's Degree in Biotechnology			2	COMPULSO		Attendance- based	
Course number of credits (ECTS)	6							
Type of activity, credits, and groups	Activity type	PRA	LAB	Р	RAULA		TEORIA	
	Number of credits	0.5	0.3		2.2		3	
	Number of groups	3	5		2		1	
Coordination	GARI MARSOL, ELOI							
Department	BASIC MEDICAL SCIENCES							
Teaching load distribution between lectures and independent student work	Hours teaching 60 Hours studying 90 Master classes 30 hours Lab practices 8 hours Practical course 22 hours							
Important information on data processing	Consult this link for more information.							
Language	Catalan for teaching English for protocols Catalan, Spanish and English for questions and tutoring							
Distribution of credits	50% theory 50% practice							

Teaching staff	E-mail addresses	Credits taught by teacher	Office and hour of attention
FERREZUELO MUÑOZ, FRANCISCO	francisco.ferrezuelo@udl.cat	1,5	
GARI MARSOL, ELOI	eloi.gari@udl.cat	8,9	

Subject's extra information

Context in education

The topic of genetic engineering introduces students to the knowledge of the techniques and methods for the isolation, amplification and manipulation of genes. The main objective is the application in model organisms for research. This course is mandatory and is located in the 2nd year of the degree of biotechnology, once the student has acquired the basic knowledge of cell biology and molecular genetics and microbiology. Later, in the itinerary at the 3rd and 4th years, students study theory and methods of application in human samples and model organisms, improved diagnostic and quality control, and complement the knowledge acquired in the subject of genetic engineering.

Learning objectives

- Demonstrate knowledge of the basic concepts and terminology related to processes of isolation, amplification and manipulation of genes. Topics from 1 to 9. Skills 50, 51 and 60.
- Demonstrate knowledge of techniques, methodologies and basic processes required to identify, clone and manipulate genes. Topics from 1 to 9. Skills 50, 51 and 60.
- Demonstrate knowledge of the particularities of biotechnology interests of different groups of organisms, especially the most common host organisms in genetic engineering. Topics from 10 to 14. Skills 50, 51 and 60
- The student should be able to design strategies and solve simple cloning and mutagenesis. Practices and problems. Skills 57 to 60.
- The student should be able to decide between different expression systems and host organisms according to the purpose of the experimental process and / or production. Practices and problems. Skills 57 to 60.

Competences

- To know the singularities of molecular genetic analysis and its biotechnological implications.
- To Know the fundamentals and know how to apply the methodology used in the genetic modification of organisms.
- To Know how to apply and perform electrophoretic methods for separation of proteins and nucleic acids
- To know how to apply and to carry out qualitative and quantitative immunological techniques applied to the analysis of molecules and cells
- To Know how to use basic equipment for luminometry, cytometry, chromatography and spectrometry.
- To Manage and know how to apply the basic methods of Molecular Biology used in research

Subject contents

- **Topic 1. Isolation and purification of nucleic acids.** Cell lysis. Purification of DNA and RNA. Alkaline lysis. Separation of DNA fragments by electrophoresis. Purification of DNA fragments. Pulsed field. 2h
- **Topic 2. Recombinant DNA I.** Nucleic acid degradation. Nucleases. Restriction endonucleases. Nucleic acid synthesis. Polymerases. Modification of nucleic acids. Ligase. 3h
- **Topic 3- Recombinant DNA II**. Plasmids and vectors. Methods for cloning. Selection of recombinant clones. Cloning strategies. Fill-in. Linkers. Gene libraries. 2h
- **Topic 4- Polymerase Chain Reaction.** Heat resistant polymerase. Stages and PCR reaction. Design of primers. DNA polymerase features. PCR efficiency. Reverse transcriptase PCR and RT. PCR cloning. Real time PCR. Digital PCR. 4h
- Topic 5- PCR Cloning. TA cloning. Gibson Assembly. Topo cloning. Gateway cloning. Recombinant cloning. 2h
- **Topic 6- Transformation of DNA. Vectors**. Protocols of transformation. Vectors with broad spectrum of host. Transfection protocols. Lentiviral vectors. 3h
- **Topic 7- Genetic manipulation.** Minitransposons. Integration Cassettes. Gene tagging. RNAi. Knock out. KO conditional. Knock in. GMOs. Gene therapy. 2h
- **Topic 8- Expression vectors.** Features of an expression vector. Inducible promoters. Gene reporters. In vitro transcription and translation. 2h
- **Topic 9- Production of heterologous proteins.** Conditions for maximum expression and production. Codon usage. Glycosylation. Protein purification. Secretion Vectors. Production in transgenic organisms. Baculovirus and insect cells. 3h

Part 2. Molecular Biology Techniques

- Topic 10- Gene synthesis. Oligonucleotide synthesis. Gene synthesis. PCR Assembly. Genome synthesis. 2h
- **Topic 11- DNA sequencing.** Dydeoxi method. Next Generation Sequencing. Solid-Bridge and emulsion. Ion Torrent. 3h
- **Topic 12- Hybridization of nucleic acids.** Hybridization. Supports hybridization. Synthesizing probes. Conditions of hybridization. Southern Blot. Northern Blot. Microarray. FISH. 2h

Problems and examples

- **1. Quantification and structure of nucleic acids.** Calculating the concentration of nucleic acids designed for ligation reactions, phosphorylation and ends-modification. Restriction mapping, Genetic and physical maps. 6h
- **2. Cloning strategies**. Having different genes and vectors, the student have to design protocols to clone DNA. Selection of the best strategy. 6h
- **3. Design of PCR primers.** Primers for gene amplification. Calculations of the melting and anneling temperatures. PCR conditions. 6h
- 4- Strategies for site-directed mutagenesis. Learning primers design to produce different types of mutations. 4h

Practical courses

- **Practice 1- Purification of DNA fragment from an agarose gel.** Electrophoresis of digested DNA and recovery of a fragment from agarose gel. Columns of silica gel. Finally check purification. 5h
- Practice 2 Purification of RNA from human biopsies. Determination of RQI. In collaboration with the Tissue

Bank (biobank) of IRBLleida, total RNA is obtained from frozen biopsies by using the robotic system, Maxwell® 16 LEV simplyRNA Tissue Kit Promega. Finally, the students evaluate the integrity of the RNA extracted measuring the quality of RNA (RQI) with a device from BioRad Experion. 3h

Methodology

To achieve the objectives and acquire the skills will be scheduled the following activities:

- Lectures. (CM)

These will be conducted with all students

The main goal is to give an overview of the thematic content of the methodologies and techniques.

- Problems. (Pro)

This will be done in groups of 20 students maximum. We provide to the students a list of quantification, cloning, primers design and mutagenesis problems. Students must solve these problems which will serve as a model for the exam questions.

The problems have as a main objective that the students implement the technical concepts and design methodologies in real situations in the laboratory.

-Laboratory. (PL).

The labs are intended for students to relate the theoretical aspects of the methodologies with practical protocols for different products and kits used in the molecular biology laboratory. These protocols are always in English so students should have a basic level of English. These practices will be conducted in groups of 20 students maximum (first practice) or maximum 10 students (second practice).

Development plan

Theory.

Part 1. Recombinant DNA technology. Topics from 1 to 9. 23 hours. Professor Eloi Garí

Part 2. Techniques of Molecular Biology. Topics from 10 to 12. 7 hours. Professor Eloi Garí

Problems

Quantification and structure of nucleic acids. Cloning procedures. Design of primers and PCR. Site-directed mutagenesis. 22 hours. Professor Eloi Garí. Two groups of 20-25 students.

Practices

Practice 1- Purification of a DNA fragment from an agarose gel. 5 hours. Professor Francisco Ferrezuelo. Three groups of 15 students.

Practice 2- RNA purification of human biopsies. 3 hours. Professor Eloi Garí. Five groups of 9 students.

It is MANDATORY that students bring in the course of teaching practices:

White-lab coat

Safety glasses

Chemical protection gloves

GENERAL SAFETY RULES IN LABORATORY PRACTICES

- Maintain the place of performance of clean and tidy practices. The work table must be free of backpacks, folders, coats ...
- In the laboratory you can not come with shorts or short skirts.
- Bring closed and covered shoes during the performance of the practices.
- Bring long hair always collected
- Keep the gowns cords to protect against spills and spills of chemical substances.
- Do not wear wide bracelets, pendants or sleeves that can be trapped by the equipment.
- Avoid wearing contact lenses, since the effect of chemicals is much greater if they are introduced between the contact lens and the cornea.
 - Do not eat or drink in the laboratory
 - -Smoking is forbidden within laboratories
 - -Wash your hands whenever you have contact with a chemical and before leaving the laboratory.
 - -Follow the teacher's instructions and consult any questions about security

Evaluation

There will be three exams:

Evaluation of the work in the laboratory using short questions to assess the student's ability to understand the protocols used in practice. This evaluation will represent 10% of the final grade.

Assess the knowledge of students with the theoretical concepts given in the first part of the course. There will be a multiple choice assessment for the chapters 1 up to 9th. This evaluation will represent 40% of the final grade. For chapters 10 to 12 the evaluation will be in format of short questions counting a 10% of the final score.

Finally, we will assess the ability to solve problems and cases in the field of basic genetic engineering. There will be a written assessment in the form of problem solving cases. This evaluation will represent 40% of the final grade.

To pass the course students have to obtain a global score of 60%.

Bibliography

Basic

- -"Cálculo en Biología Molecular y Biotecnología. Guía de matemáticas para el laboratorio". 2ª Edició. 2012. F.H. STEPHENSON. Academic Press. Ed. Elsevier
- "Molecular Biotechnology" 3ª Edició. 2003. BR. GLICK, JJ. PASTERNAK. ASM Press.
- -"**Principles of Gene Manipulation**" 6ª Edició. 2001. SB. PRIMROSE, RM. TWYMAN, RW. OLD. Blackwell Science Publishers.
- -"Gene Cloning and DNA analysis. An Introduction" 4ª Edició. 2001. TA. BROWN. Blackwell Science Publishers.
- -"Ingeniería Genética y Transferencía Génica" 1ª Edició. 1999. M. IZQUIERDO ROJO. Ed. Pirámide.
- -"Ingeniería Genética. Vol I i II" 1ª Edició. 2002. J. PERERA, A. TORMO i JL. GARCIA. Ed. Síntesis
- -"Tècnicas de Ingeniería Genética". 1ª Edició. 2017. MD. REAL GARCÍA, C. RAUSELL i A. LATORRE. Ed. Síntesis

Complementary

-"Current Protocols in Molecular Biology" Edició en CD rom. 2006. John Wiley Press.

- -"Molecular Cloning. A Laboratory Manual" 3ª Edició. 2001. J. SAMBROOK, DW. RUSSELL. CSHL Press.
- -"Biotechnologie" 5ª Edició. 1999. R. SCRIBAN (coord). Ed. Tec&Doc.
- "DNA Science. A First Course." 2ª Edició. 2003. DA. MICKLOS, GA FREYER. CSHL Press.
- -"Genes VIII" 8ª Edició. 2004. B. LEWIN. Prentice Hall.
- "Biología Molecular e Ingeniería Genética. Conceptos técnicas y aplicaciones en ciencias de la salud". 2ª Edició. 2012. A. HERRÀEZ. Ed. Elsevier

Adaptations to the contents due to COVID-19

Contents

We will be able to give practically all the contents, the theory classes were made almost all in person. The practices were all also done in person. We have two variations,

- 1- the last theory topics, topic 11 and 12, have not been given whole in theory classes. The concepts in these topics are incorporated into problem classes.
- 2- The contents of problems will all be given as planned, albeit in person.

Adaptations to the methodology due to COVID-19

Methodology

Problem and case sessions will all be held, 11 two-hour sessions, in person by video conference. They will not be in a medium group but in a large group (the whole class). Students have the opportunity to ask questions and doubts, in addition the video conference is recorded and students can listen to it again outside class hours.

As for tutorials, they are first contacted by email to set a time, and then students can connect through campus by video conference and are attended to.

Adaptations to the development plan due to COVID-19

Development plan

As I was referring to content, some concepts of topics 11 and 12 have been incorporated into the problem sessions.

Adaptations to the evaluation due to COVID-19

Evaluations

The evaluation of the 1st part of theory and practices has had the following changes:

- The exam has been done non-face-to-face through the test tool. Everything has been done as a test type.
- the practices have not been done in a short question if not a test type.
- Theory and practice have been combined considering 50% of the evaluation.

The evaluation of the second part will be of topic 10 of theory and problems. It will be non-face-to.face, the theory in test type format. For problems it will include test type and short questions as planned. It will be 50% of the evaluation.

For the exams, it was done in the 1st part and it will be done in the second, a small non-face-to-face test exam is done before the part to see that all the students have the possibility to do it. It also serves to see what drawbacks

they have or the evaluation with the chosen format.

The final recovery exam will be tried in person, in case it is not possible, the same strategy will be implemented as in the two partial ones.