Universitat de Lleida

DEGREE CURRICULUM

GENETIC ENGINEERING

Coordination: GARÍ MARSOL, ELOI

Academic year 2020-21

GENETIC ENGINEERING 2020-21

Subject's general information

<table>
<tr><td>Subject name</td><td>GENETIC ENGINEERING</td></tr>
<tr><td>Code</td><td>101611</td></tr>
<tr><td>Semester</td><td>2nd Q(SEMESTER) CONTINUED EVALUATION</td></tr>
<tr><td>Typology</td><td>
<table>
<tr><th>Degree</th><th>Course</th><th>Character</th><th>Modality</th></tr>
<tr><td>Bachelor's Degree in Biotechnology</td><td>2</td><td>COMPULSORY</td><td>Attendance-based</td></tr>
</table>
</td></tr>
<tr><td>Course number of credits (ECTS)</td><td>6</td></tr>
<tr><td>Type of activity, credits, and groups</td><td>
<table>
<tr><th>Activity type</th><th colspan="2">PRALAB</th><th>PRAULA</th><th>TEORIA</th></tr>
<tr><td>Number of credits</td><td>0.5</td><td>0.3</td><td>2.2</td><td>3</td></tr>
<tr><td>Number of groups</td><td>3</td><td>5</td><td>2</td><td>1</td></tr>
</table>
</td></tr>
<tr><td>Coordination</td><td>GARÍ MARSOL, ELOI</td></tr>
<tr><td>Department</td><td>BASIC MEDICAL SCIENCES</td></tr>
<tr><td>Teaching load distribution between lectures and independent student work</td><td>Hours teaching 60
Hours studying 90
Master classes 30 hours
Lab practices 5 hours
Practical course 25 hours</td></tr>
<tr><td>Important information on data processing</td><td>Consult this link for more information.</td></tr>
<tr><td>Language</td><td>Catalan for teaching
English for protocols
Catalan, Spanish and English for questions and tutoring</td></tr>
<tr><td>Distribution of credits</td><td>50% theory
50% practice</td></tr>
</table>

GENETIC ENGINEERING 2020-21

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

Subject's extra information

Additional information in the teaching guide depending on how the covid-19 pandemic evolves. In case the severity of the pandemic persists, the subject will be done part-time with the following changes:

1- the contents, objectives and competencies are not altered. If the teaching methodology will be changed in some cases.

2- The 30 hours of master classes will be given mostly by videoconference where students can ask questions and participate. Classes will be recorded so that students have later access to the contents.

3- The 22 hours of problems and cases are prepared to be done by videoconference, they will try to do it in person in two groups as there is availability of classrooms.

4- The practices will be done in person, from 8 hours to 5 hours and the number of groups is increased to have the minimum number of students in the lab. The 3 canceled hours correspond to practice 2 which is carried out at the IRBLleida biobank and which, due to biosecurity reasons, will not be accessible during this academic year. These contents are incorporated into problems and cases.

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

Part 1. DNA cloning

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). **This practice is carried out in the Biobank of the IRBLleida in small groups, but due to the pandemic and biosafety problems this course is canceled. The contents are incorporated in 3 specific hours within quantification and structure of nucleic acids.**

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)

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 small groups.

Development plan

Theory. Mostly videoconferences

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. Semi-attended

Quantification and structure of nucleic acids. Cloning procedures. Design of primers and PCR. Site-directed mutagenesis. 22 hours. Professor Eloi Garí. Two groups depending on classroms availability.

Practices. Attended

Practice 1- Purification of a DNA fragment from an agarose gel. 5 hours. Professor Eloi Garí. Five groups of 8 students.

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

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