COURSE DATA

DATA SUBJECT

Code: 33139
Name: Genetic engineering
Cycle: Undergraduate Studies
ECTS Credits: 6
Academic year: 2026-27

STUDY (S)

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

SUBJECT-MATTER

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

COORDINATION

ESTRUCH ROS FRANCISCO

SUMMARY

The objective of this subject is to provide the basic theoretical knowledge about the methods used in Molecular Biology and Recombinant DNA Technology and their applications to the areas of Animal and Human Biology. The practical part of the course aims to familiarize students with the techniques most frequently used in laboratories of Molecular Biology and Genetic Engineering.

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

0. Introduction.

Genetic Engineering: definitions. Genetic Engineering Tools. Recombinant DNA. Historical development. Responsible use of Genetic Engineering.

1. Basic enzymology used in the manipulation of nucleic acids.

Specific nucleases: Type IIP and type S restriction enzymes. DNA polymerases: DNA pol I and the Klenow fragment, reverse transcriptase, thermostable polymerases. DNA ligases. Non-specific nucleases. Polynucleotide kinase. Alkaline phosphatase.

2. Obtaining, purification and characterization of nucleic acids.

Rapid methods for obtaining DNA. Standard nucleic acid extraction procedure. Purification and/or concentration of nucleic acids. Obtaining plasmids. Quantitative and qualitative analysis of nucleic acids. Identification of specific nucleic acid sequences in complex mixtures: obtaining probes and hybridization techniques. Southern Blot and Northern Blot.

3. Basic cloning vectors.

Plasmids: types and characteristics. Alpha complementation. Bacteriophages. Phagemids. Cosmids. Artificial chromosomes. Bi-functional vectors or shuttle. Yeast plasmids. Transformation of bacteria and yeast. Vectors and procedures for the heterologous expression of proteins.

4. Synthesis of specific DNA fragments.

cDNA synthesis. Obtaining complete cDNAs. Oligonucleotide synthesis. The polymerase chain reaction (PCR). Types of PCR. Isothermal amplification of nucleic acids.

5. Cloning strategies.

Cloning of restriction fragments. Generation and union of restriction fragments. Generation of blunt and protruding ends. Non-directional and directional cloning. Complete and partial restrictions. Construction and characterization of genomic DNA libraries. Cloning of cDNAs. cDNA libraries. Cloning of PCR products. TA cloning. Addition of sequences at the ends of a PCR fragment. Special cloning techniques: User assembly. Gibson assembly. Golden Gate. Gateway cloning. Cloning by recombination: cloning by homologous recombination in yeast: TAR (transformation-associated recombination) cloning. Synthetic genomics.

6. DNA sequencing.

Sanger's method. Obtaining the sequence of complete genomes: the reference genomes. New high throughput sequencing (NGS) methods: second and third generation.

7. Techniques for the study of gene expression.

Northern blot. DNA arrays. Quantification of transcripts by PCR. semiquantitative PCR. real-time PCR. RNA-seq. RNA-seq of individual cells.

8. Identification and characterization of physical interactions between macromolecules.

Protein-protein interactions: Protein co-immunoprecipitation. The TAP technique. The double hybrid method. Protein-DNA interactions: Mobility changes in gel. Chromatin immunoprecipitation (ChIp).

9. Modification of the DNA sequence

Forward genetics and reverse genetics. Random mutation. Chemical mutagenesis. Identification of mutants and allelic classification in yeast. Cloning by complementation. Insertional mutagenesis in yeast and mice. Genetic interactions between mutant alleles: suppression and synthetic lethality. Specific changes in the sequence. Directed mutagenesis in vitro. Genetic disruptions. PCR-based targeted mutagenesis techniques. Exchange of wild sequences for mutated sequences in vitro through homologous recombination. Genomic addition of tags to proteins. Directed evolution and phage display.

10. Genetic modification in cells and higher organisms.

Transfection and transduction. Transitory and permanent expression. Main viral vectors. Adenovirus. Adeno-associated viruses. Retroviruses and lentiviruses. Genome editing in complex organisms. High specificity nucleases. Meganucleases. Zinc finger nucleases. TALEN. Cas9/CRISPR. Gene therapy: CAR-T. Gene therapy: SMA and sickle cell anemia. Gene drive. Massive mutagenesis in higher cells and phenotypic identification of mutants. Synthetic lethality and cancer.

11. Transgenic animals.

Construction and applications. Manipulation of embryonic cells. Microinjection. Transfection tests. Obtaining cloned animals.

12. LABORATORY EXPERIENCES

Practice 1. - Construction of a library.
Practice 2. - Protein expression in Escherichia coli.

Practice 3. - Construction of the restriction map of a plasmid.
Practice 4. - Detection and characterization of transgenic yeast.
Practice 5 - Identification of interactions between proteins using the two-hybrid method

WORKLOAD

PRESENCIAL ACTIVITIES

Activity	Hours
Theory	40,00
Laboratory	16,00
Classroom practices	4,00
Total hours	**60,00**

NON PRESENCIAL ACTIVITIES

Activity	Hours
Attendance at other activities	0,00
Individual or group project	0,00
Independent study and work	0,00
Preparation of lessons	90,00
Preparation for assessment activities	0,00
Resolution of case studies	0,00
Total hours	**90,00**

TEACHING METHODOLOGY

The development of the course is structured around theoretical sessions, personal coaching or through e-mail.

Theoretical sessions: A total of 35 one-hour classroom sessions for lectures and 4 one-hour of question resolving will be scheduled. Before each lesson, students will be provided with all significant artwork that will be presented. This material will be available in multimedia. Thus, it is intended that the student can follow the explanations more easily, taking only the notes needed for proper understanding.

Personal tutoring: The role of tutoring is to help and guide the student personally in all the problems that arise in dealing with the study of the subject, and facilitate the exchange of views between teacher and student, in an effort to approach teaching in a personalized way. Communication on-line can also be used to enhance teacher-student interaction. In any case, preferred tutorial classes are much more suitable for the explanation of the doubts and questions.

Practical activities: 5 sessions are scheduled from 2 to 4 hours each (total 17 h) on various aspects of molecular biology techniques and recombinant DNA that are adaptable to laboratory practice.

EVALUATION

At the end of the course, an examination will be conducted to evaluate the knowledge acquired in theoretical and practical classes.

The theoretical part will have a value of 85% of the final grade while practices account for 15% (distributed between the lab report, 7.5%, and one or more questions related to practices that will be included in the final exam, which represent 7.5% of the note). To pass the course, it will be needed a minimum score of 4.5 points from the 9,25 points of the exam (8.5 points from theory and 0.75 points from the questions related with practices).

The voluntary submission of responses to the questions raised by the teacher during tutoring sessions will be further assessed with up to 0.5 points.

Attendance at practices is mandatory and so is the presentation of the lab report. In case of failure to pass the course, the score corresponding to the report and the questions related with practices of the exam can be kept for the following call or course.

REFERENCES

En este apartado se incluyen únicamente aquellos textos que se pueden considerar como de uso general en la asignatura, con un comentario crítico de cada uno de ellos.

Se ha considerado conveniente dividir los textos generales en dos grupos.

Libros de texto. Estos libros están escritos para explicar las metodologías desde un punto de vista docente y bastante teórico. Son lo más apropiados para la asignatura.

BROWN, T.A. (2020). Gene cloning and DNA analysis. An introduction. 8ª edición. Ed. Blackwell Science. Un excelente libro de texto de un nivel más sencillo que otros, pero muy bien explicado y actualizado.

REAL GARCÍA, M. D.; RAUSELL SEGARRA, C. Y LATORRE CASTILLO, A. (2017). Técnicas de ingeniería genética. Editorial Síntesis. Un texto en castellano que trata de forma detallada las principales técnicas de la Ingeniería Genética

LEWIN, B. (2010). "Genes X". Jones & Bartlett. Es el libro más conocido como texto de Biología Molecular. Para Métodos no es muy apropiado pero, aún así, tiene algunos capítulos de utilidad.

LUQUE, J. y HERRAEZ, A. (2001) Biología Molecular e Ingeniería Genética. Harcourt. Muy amplio de contenidos pero tiene buenos esquemas para algunos capítulos de esta asignatura.

Libros de protocolos. Estos libros han sido escritos para el uso de los investigadores en los laboratorios. Contienen recetas de los distintos protocolos así como escuetas explicaciones teóricas de los fundamentos de los métodos y posibles artefactos. No son muy apropiados para aprender los principios

básicos de las metodologías pero pueden ayudar a aclarar algunos puntos concretos y conviene familiarizarse con ellos pues son los que se usarán después en el laboratorio.

AUSUBEL, F.M. et al. (1987-97). ¿Current protocols in Molecular Biology¿. John Wiley & sons. Quizá el libro más completo de protocolos. Además, debido a su formato de hojas recambiables, se actualiza de forma periódica.

BIRREN ET AL. (1999). Genome analysis. 4 Volúmenes. Cold Spring Harb. Lab. Press. Uno de los más completos libros de protocolos, considerando los 4 volúmenes. Incluye aplicaciones de metodología de Ingeniería Genética.

SAMBROOK, J. y RUSSELL, D.W. (2001). ¿Molecular Cloning. A laboratory manual¿. 3ª ed. Cold Spring Harbor Laboratory Press (3 volúmenes). El más famoso libro de protocolos de Biología Molecular. Es uno de los más populares y la actualización del mismo es muy reciente.

DIEFFENBACH, C.W. y DVEKSLER, G.S. (1995). PCR primer. A laboratory manual. Cold Spring Harbor. Un manual específico sobre PCR.