Universitat Autònoma de Barcelona

2021/2022

Genetic Engineering of Microorganisms

Code: 100972
ECTS Credits: 6

Degree	Type	Year	Semester
2500253 Biotechnology	OT	4	0

The proposed teaching and assessment methodology that appear in the guide may be subject to changes as a result of the restrictions to face-to-face class attendance imposed by the health authorities.

Contact

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Use of Languages

Principal working language: spanish (spa)
Some groups entirely in English: No
Some groups entirely in Catalan: No
Some groups entirely in Spanish: Yes

Other comments on languages

We will work with material in English.

Teachers

Jesús Aranda Rodríguez

Prerequisites

It is recommendable to have studied or are studying Microbiology, Genetics, Molecular Biology and Genetics, Molecular Microbiology and Virology.

Objectives and Contextualisation

The main objective of this course is that the student will be able to design procedures for the genetic manipulation of microorganisms.

Therefore during the development of the subject, the student must reach the following capacities:
• To know how to identify different types of microbial vectors, recognize their applications and design new ones
• To know how to apply methodologies and strategies of cloning
• To recognize the implication of the characteristics of each microorganism (immunity systems, recombination capacity, codon usage, etc.) in the proposed experimental design
• To know how to choose the most appropriate genetic transfer technique in each proposed case
• To be able to design efficient strategies for obtaining, enriching and selecting mutants
• To know how to build gene fusions and recognize their possible applications
• To recognize the main characteristics of potential bacterial targets for drugs, vaccines, and diagnostic reagents development.

Competences

- Apply the principal techniques for the use of biological systems: recombinant DNA and cloning, cell cultures, manipulation of viruses, bacteria and animal and plant cells, immunological techniques, microscopy techniques, recombinant proteins and methods of separation and characterisation of biomolecules.
- Comply with ethical principles and legislation in the manipulation of biological systems.
- Identify the genetic, physiological and metabolic properties of microorganisms with potential for application to biotechnological processes and the possibility of manipulating microorganisms.
- Interpret experimental results and identify consistent and inconsistent elements.
- Read specialised texts both in English and ones own language.
- Reason in a critical manner
- Search for and manage information from various sources.
- Think in an integrated manner and approach problems from different perspectives.
- Work individually and in teams

Learning Outcomes

1. Comply with ethical principles and legislation in the manipulation of of microorganisms.
2. Describe the principal techniques associated with the genetic manipulation of microorganisms.
3. Identify the potential for manipulation of microorganisms.
4. Interpret experimental results and identify consistent and inconsistent elements.
5. Read specialised texts both in English and ones own language.
6. Reason in a critical manner
7. Search for and manage information from various sources.
8. Think in an integrated manner and approach problems from different perspectives.
9. Work individually and in teams

Content

The content of the course consists of the following topics*:

Unit 1. DNA introduction systems in bacteria. Natural transformation in gramnegative and grampositive bacteria. State of competence. Molecular mechanisms associated with natural transformation. Induced transformation. Electrotransformation. Design and optimization of transformation systems in various bacteria lacking natural transformation.

Unit 2. DNA vectors in bacteria. Requirements of cloning vectors. Expression vectors. T-type vectors. Mobilizable vectors. Suicide vectors. Shuttle vectors. Integrational vectors. Genetic characteristics of vector accepting cells. Construction of DNA libraries *in vitro* and *in vivo*.

Unit 3. Bacterial gene fusions. Transcriptional and translational fusions. Gene fusions in polycistronic units. Fusion vectors: general characteristics. Random gene fusions. Methods for the construction of gene fusions. Construction of gene fusions by PCR, OE-PCR and Gibson assembly. Applications and examples of gene fusions.

Unit 4. Mutagenesis in bacteria. Random mutagenesis *in vivo*. Use of chemical or physical methods. Criteria and methods for the selection and enrichment of mutants. Transposons. Minitransposons. Plasposons. Transposomes. Methods for the identification and confirmation of mutants. *In vitro* mutagenesis of cloned genes.

Unit 5. Cloning strategies in bacteria. Cloning by complementation: anabolic or catabolic genes. Regulatory gene isolation methods. Cloning of toxic genes. Obtaining virulence genes.

Unit 6. Gene substitution in bacteria and generation of knockouts. Obtaining mutants by gene disruption and by gene substitution. Lambda Red system. Obtaining scarless mutants. Counter selection systems. I-SceI system. Use of CRISPR/Cas9 technology to obtain mutants. Methods for the identification and confirmation of mutants. Systems for the reintroduction of altered genes in the bacterium of origin. Insertion into the chromosome of new genes or constructs.

*Unless the requirements enforced by the health authorities demand a prioritization or reduction of these contents.

Methodology

Genetic Engineering of Prokaryotes course is organized in two modules:

Theoretical module: where participatory masterclasses are combined with problem-based learning sessions where theoretical concepts are worked through the resolution of practical cases.

Seminar module: in which through collaborative learning, students work on different aspects of actual experimental designs present in recent scientific articles. At the beginning of the course, students choose, following the guidelines set by the teaching staff, a scientific article related to the field of genetic engineering of microorganisms from which they make a poster. The schedule of activities like classroom work sessions, exhibition, and discussions, as well as the delivery dates of the proposed activities, will be defined at the beginning of the course by the teachers.

(The proposed teaching methodology may experience some modifications depending on the restrictions to face-to-face activities enforced by health authorities.)

Annotation: Within the schedule set by the centre or degree programme, 15 minutes of one class will be reserved for students to evaluate their lecturers and their courses or modules through questionnaires.

Activities

Title	Hours	ECTS	Learning Outcomes
Type: Directed			
Participatory master classes	30	1.2	1, 2, 3, 6
Seminars	12	0.48	1, 7, 4, 5, 8, 6, 9
Type: Supervised			
Tutorship	1	0.04	
Type: Autonomous			
Preparation of posters and questionnaires	34	1.36	1, 7, 3, 4, 5, 8, 6, 9
Reading recommended texts	20	0.8	5
Study and other autolearning activities	50	2	1, 7, 3, 4, 5, 8, 6, 9

Assessment

Seminar module evaluation
The evaluation of the seminars is done through the evaluation of different activities related to a scientific article:

A) Autonomous deliveries that will be delivered through the Moodle classroom and deliveries in the classroom work sessions. With a maximum rating of 2 points out of 10.

B) The poster and questionnaire associated with the chosen scientific article. With a maximum rating of 5 points out of 10.

C) The defense of the poster during its classroom exhibition. With a maximum rating of 1 point out of 10.

D) The resolution of the questionnaires related to the presented seminars. With a maximum rating of 1.5 points out of 10.

E) Individual and workgroup self-evaluation. With a maximum rating of 0.5 points out of 10.

To pass this module the student must obtain a grade equal or superior to 5.

Theoretical module evaluation
The evaluation of this activity is done through an individual written exam. The maximum rating of this section is 10 points out of 10.
To pass this module it is necessary to obtain a score equal to or greater than 5 points.

If the grade obtained is less than 5, the student must take the retake examination. This test will have a maximum qualification of 8 points out of 10 and a score equal to or greater than 4 will be necessary to pass the module.

To be eligible for the retake process, the student should have been previously evaluated in a set of activities equaling at least two-thirds of the final score of the course or module.

Students who have passed the module may submit to a grade improvement test waiving the grade obtained previously in the individual written exam. The scheduled date for the second chance test is that of the second chance examination. Students wishing to take the grade improvement test must communicate it by mail to the teacher responsible for the subject at least 72 hours before the day scheduled for the second chance examination.

The final grade of the course will be the average of the grades obtained in both modules, being necessary to have passed separately each of them.
The student will be graded as "Non-evaluable" if the weight in of all conducted evaluation activities is less than 67% of the final score.

Student's assessment may experience some modifications depending on the restrictions to face-to-face activities enforced by health authorities.

Assessment Activities

Title	Weighting	Hours	ECTS	Learning Outcomes
Classroom and virtual classroom submissions	10%	0	0	7, 5, 8, 6, 9
Discussion and participation in the classroom	5%	0	0	1, 2, 3, 4, 8, 6
Poster	25%	0	0	1, 7, 2, 3, 4, 5, 8, 6, 9
Resolution of questionnaires in the classroom	7.5%	0	0	1, 2, 3, 4, 8, 6
Written test (resolution of practical cases)	50%	3	0.12	2, 3, 4, 8, 6, 9
team-work or individual self-evaluation	2.5%	0	0	1, 7, 2, 3, 4, 5, 8, 6, 9

Bibliography

As reference bibliography of basic concepts it is recommended:

Snyder, Larry. Molecular Genetics of Bacteria. 4th ed. Washington, DC: ASM Press. 2013.

eBook available at Biblioteques UAB:

http://eds.b.ebscohost.com/eds/detail/detail?vid=0&sid=aaef5fab-a6ed-47c8-9dc3-70a5856ae074%40sessionmg

Jeremy W. Dale, Simon F. Park. Molecular Genetics of Bacteria. 5th ed. Chichester, West Sussex, England : Wiley. 2010.

eBook available at Biblioteques UAB:

http://eds.b.ebscohost.com/eds/detail/detail?vid=0&sid=f3365a86-b2bc-4182-ac49-58f4d3bf00ca%40pdc-v-sessi

Other recommended texts as well as links of interest will be available to the student in the Moodle classroom of the course.

Software

Not applicable.