Universitat Autònoma de Barcelona

2018/2019

Laboratory IV

Code: 100977
ECTS Credits: 3

Degree	Type	Year	Semester
2500502 Microbiology	OB	2	2

Contact

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Use of languages

Principal working language: catalan (cat)
Some groups entirely in English: No
Some groups entirely in Catalan: Yes
Some groups entirely in Spanish: No

Teachers

Neus Ferrer Miralles
Eduard Villagrasa Ramirez
Concepcion de Linares Fernandez
Nuria Vigues Frantzen
Maria Constenla Matalobos

Prerequisites

Students are advised to review the scientific-technical content on which this subject is based.

It would be desirable if students review basic concepts of the microbial world previously acquired in the first course of the Degree in Microbiology, especially the subjects of *Laboratoris integrats I* and *II*, and the subjects programmed in the first semester of the second course, such as *Laboratori integrat III* and Protistology, and also a good knowledge about the subjects coursed simultaneously in the second semester of the second course.

Students must have passed the biosafety and security laboratory tests to attend this course, and be knowledgeable and accept the laboratories regulations of the *Facultat de Biociències*.

Objectives and Contextualisation

This is a compulsory subject, a nuclear course from the degree of Microbiology, which introduces students to the knowledge of different microbiological techniques to study in situ the ecophysiology in natural or artificial environments and in a microbiology laboratory. The achievement of the competencies of the course will allow students acquire new knowledge related to other practical and theoretical subjects simultaneously and subsequently coursed in the degree of Microbiology.

The main objectives are:

1. Know and use different methodologies to study the microbial ecosystems physiology.

2. Elaborate and analysis of experimental models of the laboratory.
3. Apply methodologies related to counting and isolation of microorganisms, biomass determinations, microbial activity and primary production measurements
4. Integration of methodologies for the microbial community characterization (prokaryotes and eukaryotes) that form part of a specific ecosystem.

Skills

- Apply knowledge of theory to practice
- Apply suitable methodologies for taking samples and characterising and manipulating microbial populations and communities in natural and artificial ecosystems, and establish the relationships between these and those with other organisms.
- Apply suitable methodologies to isolate, analyse, observe, cultivate, identify and conserve microorganisms.
- Apply the principles of risk assessment and prevention in the laboratory, and biosafety regulations on microorganisms and manipulation of different biological systems.
- Characterise the causal agents of microbial diseases in humans, animals and plants in order to diagnose and control them, perform epidemiological studies and be aware of present-day problems with these diseases and strategies to combat them.
- Communicate orally and in writing.
- Display sensibility towards environmental, health and social matters.
- Identify and solve problems.
- Obtain, select and manage information.
- Use bibliography or internet tools, specific to microbiology or other related disciplines, both in English and in the first language.
- Work individually or in groups, in multidisciplinary teams and in an international context.

Learning outcomes

1. Apply general procedures for enriching and selecting microorganisms.
2. Apply in the laboratory the principles of risk assessment and prevention.
3. Apply knowledge of theory to practice
4. Apply methods for processing samples for subsequent microbiological analysis.
5. Apply strategies for selecting and enriching different functional groups of microorganisms.
6. Apply suitable sampling strategies and techniques for different types of environments.
7. Characterise environmental parameters and relate them to the habitat of the different functional groups of microorganisms.
8. Communicate orally and in writing.
9. Create microcosms and reproduce natural environments in the laboratory.
10. Determine microbial concentration in several samples by using microscopy and seeding methods.
11. Display sensibility towards environmental, health and social matters.
12. Evaluate and interpret microbial activity in natural environments.
13. Identify and solve problems.
14. Interpret electron microscope images related to microorganisms.
15. Obtain, select and manage information.
16. Solve basic calculus problems in quantitative microbiology.
17. Use advanced techniques for seeding and culturing microorganisms.
18. Use bibliography or internet tools, specific to microbiology or other related disciplines, both in English and in the first language.
19. Use conventional techniques for identifying microorganisms.
20. Use different methods for characterising microbial communities.
21. Use electron microscopy techniques to visualise viruses in a sample.
22. Use suitable methodologies for determining the concentration of viruses in a sample.
23. Work individually or in groups, in multidisciplinary teams and in an international context.

Content

The subject is organized into three sections, as detailed below:

Section 1: Methodological block

3 methodological and monographic sessions of three hours each session.

Section 2: Characterization of experimental models of laboratory (microcosms)

7 sessions related to the characterization of the microcosms designed in the previous module. These sessions are divided into 2 different weeks. The first week corresponds to 3 sessions of three hours and a session of four hours, and the second week with 2 sessions of three hours and a sessió of two hours.

Section 3: Study of a natural environment: microbial diversity

In this module, a field trip to a specific natural environment of 8 hours is programmed. Later there will be 3 sessions of 3 hours and 1 session of 2 hours, in order to analyze the field samples of microbial diversity.

The content for each module session are as follows:

Section 1: Methodological block

It is intended to introduce the student in different methodologies to study the ecophysiology of microbial ecosystems.

Session	Content
1	Phototrophic and heterotrophic microbial activity evaluation. Oxygen and sulfide: initial measures. Phototrophic (pigments) and total (DNA) microbial biomass determination.
2	Phototrophic (pigments) and total (proteins) microbial biomass determination Preparation of experimental models of laboratory (microcosms).
3	Phototrophic and heterotrophic microbial activity evaluation. Oxygen and sulfide: final measures. Primary production in planktonic environments.

Section 2: Characterization of experimental models of laboratory (microcosms)

The purpose of this module is an integration of different methodologies for the characterization of microbial communities and shows the usefulness of experimental models of the laboratory in research.

Session	Content
1	Observation and description of the experimental models. Preparation of enrichment cultures (liquids and solids) of phototrophic and heterotrophic and aerobic and anaerobic.

	Enrichment cultures of prokaryotes viruses. Plate counts of water and sediment samples: spread on plates.
2	Metabolic characterization of the microcosms (carbon sources, enzymes). Enrichment observation. Viable plate counts. Isolation and quantification of the virus: the plaque assay (part I) Isolation and quantification of a microorganism (Gram stain, cultures)
3	Metabolic characterization of the microcosms (enzymes) Quantification of the virus: the plaque assay (part II) Isolation and quantification of a microorganism (biochemical and physiological characterization). DNA extraction of the isolated microorganism from the microcosm
4	Biochemical identification of the isolated microorganism Metabolic characterization of the microcosms (carbon sources) Virus DNA extraction from the microcosm Discussion of the obtained results
5	Enrichments (observation) DNA amplification (rRNA 16S gene and viral DNA)
6	Enrichments (observation) TEM preparation of virus enrichment, PCR gel Results analysis
7	Sequencing sample preparation Viral observation by TEM Discussion

Section 3: Study of a natural environment: microbial diversity

This module takes into account the contents of the modules made previously so that all the knowledge and content achieved previously will be applied to the study of a natural ecosystem.

Session	Content
Field trip	Limnology and sampling collection techniques Physical and chemical parameters determination
1	Photosynthetic protist observation

2	Heterotrophic protist observation
3	The abundance of different bacterial groups using techniques of hybridization in situ with fluorochromes-labeled probes. Gel agarose for detection of the virus of eukaryotes. Detection of the virus of eukaryotes
4	Microbial diversity by Confocal laser scanning microscopy (CLSM). Results analysis Final discussion

Methodology

This course will be taught in three small groups of students (maximum of 24 students per session).

The class attendance is a mandatory requirement in order to pass the course. Student who cannot attend an individual or different sessions for justified and unforeseeable cause (as a health problem, death of a relative up to second degree, accident, enjoy the status of elite athlete and have a competition or sport activity of obligatory attendance, etc) shall be present, as soon as possible, official documentation to the responsible of the course (official medical certificate stating explicitly the inability to attend the session/s, police attestation, justification of the competent sports authority, etc.).

The field trip that is part of module 3 will be held jointly by all the groups of practices.

Before the beginning of the practice sessions, the students will have the Manual of the course.

At each lab session, it is compulsory for the students to wear their own lab coat, laboratory glasses, permanent marker, lighter, calculator and the Manual of the course, which will be available in the Virtual Campus, Moodle platform, or where the teacher says. It is also necessary, and only during the module 2, to carry a Miquelrius, Abacus, or Oxford notebook with sewn sheets. In this notebook lab, students must write the information according to the *Guia de característiques i anotacions* that they will find in the Manual of the course and that student should be read before starting the module 2.

Students will work in pairs and under the supervision of the teacher. At the beginning and/or during each session the teacher will make brief theoretical explanations of the practice content and procedures to be carried out by the students, as well as specific safety measures and the different chemical and biological generated waste treatment.

It is essential that the student makes a comprehensive reading of the Manualof the course, and becoming familiar with the practices that will take place in each session, as well as the methodology will be applied in each case. During each practice session, and whenever the teacher considers it necessary, it will do questions to students in reference to the previous reading of the Manual.

Activities

Title	Hours	ECTS	Learning outcomes
Type: Directed			
Field trip	8	0.32	3, 6, 4, 1, 7, 15, 8, 23, 11, 18, 20, 19, 12

Laboratory Classes	42	1.68	2, 3, 5, 6, 4, 1, 7, 9, 10, 13, 14, 15, 16, 8, 23, 11, 18, 20, 22, 21, 17, 19, 12
Presentation of the subject	1	0.04	3, 15
Type: Supervised			
Tutory	1	0.04	2, 3, 5, 6, 4, 1, 7, 9, 10, 13, 14, 15, 16, 8, 23, 11, 18, 20, 22, 21, 17, 19, 12
Type: Autonomous			
Comprehensive reading of the Manual of the course	6	0.24	3, 5, 6, 4, 1, 7, 9, 10, 14, 16, 8, 23, 11, 20, 22, 21, 17, 19, 12
Drafting of the lab delivery	2	0.08	13, 15, 8
Solving problems	2	0.08	3, 10, 13, 14, 16, 8, 23, 11, 22, 12
Study	10	0.4	2, 5, 6, 4, 1, 7, 9, 10, 14, 16, 20, 22, 21, 17, 19, 12

Evaluation

The assessment of the course will be done by section and in a continuous manner. The weight of each module respect to the final grade of the course is Section 1, 25 %; Section 2, 40 %; and Section 3, 35 %. To pass the course students must get at least a 5 in each module and in the final grade of the course.

The evaluation in each section will be as follows:

Section 1: Methodological block. Exercises delivery (5 %); Questionnaire with multiple choice questions (20 %).

Section 2: Characterization of experimental models of laboratory (microcosms). Exercises delivery (4 %); Notebook lab delivery (8 %); Questionnaire with multiple choice questions (28 %).

Section 3: Study of a natural environment: microbial diversity. This integrated section is taught by three teaching units: Microbiology (M), Botany (B) and Zoology (Z). A Visum examination (14 %, B, and Z); Report delivery (3.5 %, M); Questionnaire with multiple choice and/or short questions (17.5 %, M, B, and Z). Questions concerning the content included in the Manual of the course explained in practical laboratory sessions and/or explained in the field trip (module 3) could be included in the three questionnaires. The final grade for this module will be distributed in the following manner: Microbiology (17.5 %), Botany (8.75 %) and Zoology (8.75 %). The requirements to sum the obtained notes in the other units are A minimum note of 4.5 in each Visum examination (B and Z), and a 4 in the report delivery (M) and in the questionnaires (M, B, and Z).

In each section, the student's attitude in the laboratory, punctuality, the use of laboratory equipment (gown and goggles), compliance with the safety and biosecurity regulations, and the understanding and follow-up of the Manual of the subject will be taken into account. This assessment does not entail an increase in the score, but it can mean the reduction of up to 20% of the final mark obtained in each section.

Since attendance to the activities programmed in this course is mandatory (practical sessions and field trip), the absence of any of the sessions must be justified before 48h after the date of the activity. The non-attendance to the field trip without justified cause will result in a penalty (2 points) in the final note of Section 3, and this penalty shall be maintained in the recovery exam. In case of a strike, if a student decides to exercise his right to strike, he will have to communicate to the responsible for a maximum period of 48h after the day of the strike. In any case, the absence may exceed 20 % of the scheduled activities; If this value is exceeded, the student will be qualified as Not Assessable.

Students who do not pass the evaluations of the different sections of the course will be able to recover them in the date scheduled at the end of the semester (recovery exam), performing a questionnaire with multiple

choice and/or short questions and/or a visum examination associated with the section not previously passed. In this course, there will be no consideration of improvement, neither global nor by sections.

Students who do not obtain the minimum qualification required to pass each of the sections of the integrated laboratory, will not be able to pass the subject. In this case, the final maximum grade of the subject will be 4.

As this course is differentiated into sections, from the second enrolment, students will only have to perform again the specific sections that have not been previously passed.

Evaluation activities

Title	Weighting	Hours	ECTS	Learning outcomes
Section 1: Exercices delivery	5	0	0	3, 5, 4, 7, 10, 13, 16, 8, 23, 11, 12
Section 1: Questionnaire with multiple choice questions	20	1	0.04	2, 3, 5, 6, 4, 7, 9, 10, 13, 16, 8, 23, 11, 12
Section 2: Exercices delivery	4	0	0	3, 13, 16, 8, 23, 22, 12
Section 2: Notebook lab delivery	8	0	0	2, 3, 5, 4, 1, 7, 9, 13, 14, 15, 16, 8, 23, 11, 18, 20, 22, 17, 19, 12
Section 2: Questionnaire with multiple choice questions	28	1	0.04	2, 3, 5, 4, 1, 7, 9, 13, 14, 15, 16, 8, 23, 11, 18, 20, 22, 21, 17, 19, 12
Section 3: Questionnaire with multiple choice questions/short questions	17.5	0.5	0.02	2, 3, 6, 4, 7, 10, 13, 15, 8, 23, 11, 18, 20, 12
Section 3: Report delivery	3.5	0	0	2, 3, 6, 4, 7, 10, 13, 15, 8, 23, 11, 18, 20, 12
Section 3: Visum examination	14	0.5	0.02	2, 3, 6, 4, 7, 10, 13, 15, 8, 23, 11, 18, 20, 12

Bibliography

The objectives and the protocols that students will be used in each session will be detailed in the Manual of the course. This Manual also contains the bibliography associated with each of the different practical sessions. If it was necessary, teachers may also give further information to the students via the Virtual Campus or Moodle platform. However, some relevant references (books and web links) are listed below.

Books

Altaba, C. et al. 1991. Invertebrats no artròpodes. Història Natural dels Països Catalans. Vol. 8. Enciclopèdia Catalana. Barcelona.

Atlas, R.M. & Bartha, R. 2002. Ecologia microbiana y Microbiologia ambiental.(Trad.4a ed. americana Addison Wesley). Pearson Educación. Madrid.

Bellinger, E.G., and Sigee D.C.2010. Freshwater Algae: identification and use as bioindicators. Wiley-Blackwell.UK.

Burlage, R.S. Atlas, R., Stahl, D., Geesey, G., and Sayler, G. 1998. Techniques in microbial ecology. Oxford University Press. Washington, DC.

Cann, A. J. 2001. Principles of Molecular Virology. (3rd Ed). Academic Press.

Carrion, J. S. 2003. Evolución vegetal. DM. Murcia.

Hurst, J. 2000. Viral Ecology. Academic Press.

Llimona, X. (ed.) 1985. Plantes inferiors. Història Natural dels Països Catalans. Vol. 4. Enciclopèdia Catalana.Barcelona.

Madigan, MT., JM. Martinko, KS. Bender, DH. Buckley, DA. Stahl. 2015 (14 ed). Brock Biología de los microorganismos. Pearson Educación, S.A.

Madigan, MT., JM. Martinko, KS. Bender, DH. Buckley, DA. Stahl. 2014. Brock Biology of microorganisms.14th edition. Pearson, S.A. ISBN: 978-0-321-89739-8.

Margulis, L., Corliss, J.O., Melkonian, M, Chapman, D.J. (1990). Handbook of Protoctista. Jones& Barlett Publishers, Boston.

Maunsbach, A.B. 1998. Biomedical Electron Microscopy Illustrated Methods and Interpretations. Academic Press.

Ogunseitan, O.2005. Microbial Diversity. Blackwell Publishing.

Pepper, IL., CP. Gerba, TJ Gentry. 2014. Environmental Microbiology. 3rd edition. Academic Press (Elsevier).ISBN: 978-0-12-394626-3.

Simpson, M.G. 2006. Plant Systematics. Elsevier. Academic Press.

Streble, H. and Krauter, D. 1987. Atlas de los microorganismos de agua dulce. La vida en una gota de agua.Ed.Omega, S.A. (Barcelona).

Willey, J.M., Sherwood, L.M., and Woolverton, C.J. 2009 Microbiología de Prescott, Harley y Klein. Séptima edición. McGraw-Hill-Interamericana de España. ISBN: 978-84-481-6827-8.

Web links

Aula Virtual de l'Autònoma Interactiva: https://cv2008.uab.cat/

All the Virology on the WWW http://www.mirrorservice.org/sites/www.virology.net/

British Society for Protist Biology http://www.protist.org.uk/

Introduction to the Viruses http://www.ucmp.berkeley.edu/alllife/virus.html

Manual of Aquatic Viral Ecology http://www.aslo.org/books/mave/

Natural History Museum http://www.nhm.ac.uk/jdsml/research-uration/research/projects/protistvideo/

Tree of life web project http://tolweb.org/tree/