Biocatalysis

Code: 100956
ECTS Credits: 6

2025/2026

Degree	Type	Year
Biotechnology	OP	4

Contact

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Teaching groups languages

You can view this information at the end of this document.

Prerequisites

There are no official prerequisites. In any case, part of the contents of some subjects of 1st year and 3rd year are necessary to be able to follow the subject correctly. In particular, those of the following subjects: Biochemistry, Chemistry and Engineering of Proteins and Basic and Advanced Instrumental Techniques

Objectives and Contextualisation

Objectives and Contextualisation

The subject Biocatalysis focuses on the study of enzymes, their properties and applications. The knowledge of enzymes is key in the context of Biochemistry, Molecular Biology and related sciences, given their role as catalysts of biological reactions and their applications in biotechnological processes. The subject analyzes enzymes from different perspectives: their activity, kinetics, mechanisms and applications. The general objective of the subject is to provide the foundations for the analysis, characterization and use of enzymes from the point of view of research and from their biotechnological and biomedical applications.

Specific objectives of the subject:
Knowledge of the general characteristics, classification and testing methods of enzymatic activity.
Analysis of enzyme kinetics and determination and meaning of kinetic parameters.
Knowledge of enzyme inhibition and its applications, especially in the field of drugs.
Analysis of the active center and knowledge of the methods of characterization.
Analysis of enzymatic and regulatory mechanisms.
Biomedical and biotechnological applications of enzymes.

Use of specific software to study the structure of enzymes and modulators, as well as the enzyme kinetics.

Learning Outcomes

1. CM32 (Competence) Plan a process for obtaining biotechnological products.

2. KM34 (Knowledge) Describe the properties of microorganisms with potential application in different biotechnological processes.
3. SM33 (Skill) Interpret the kinetic parameters of enzymatic reactions, by means of graphical methods and using computer programmes.

Content

Theoretical content.

Unit 1. Introduction to biocatalysis.

Concept of biocatalysis. Market and use of biocatalysts. Prejudices in the use of enzymes. Historical perspective. Waves of innovation in biocatalysis. Advantages and disadvantages of biocatalysts. Different types of biocatalysis processes. Cellular and enzymatic systems: properties. Factors to consider in a biocatalytic process: source of the biocatalyst and optimization of the process.

Unit 2. Properties, classification and nomenclature of enzymes.

General properties of enzymes: Concept and biological, chemical and practical significance. Definitions. Enzyme-substrate complex. Decreased activation energy. Transition state. Enzymatic cofactors. Nomenclature and classification of enzymes. Databases with enzyme information.

Unit 3. Methods of determination of enzymatic activity and of obtaining enzymes.

Production and characterization of enzymes. Sources of enzymes. Techniques for the extraction of enzymes. Methods of determination of enzymatic activity. Dirct and indirect, continous and discontinous assays. Initial rate: concept, determination, representation. Units of enzymatic activity. Effect of enzyme concentration.

Unit 4. Analysis of enzyme kinetics.

Enzyme kinetics. Reactions with one substrate. Effect of substrate concentration: Michaelis-Menten equation. Pre-steady and steady-states: concepts. Steady state hypothesis: treatment of Briggs-Haldane. Enzymatic reactions with more than one enzyme-substrate intermediate complex. Significance of the parameters kcat, KM and kcat/KM. Determination of kinetic parameters. Methods with linear representations: Lineweaver-Burk, Eadie-Hofstee and Hanes-Woolf. Other methods. Michaelis-Menten equation for reversible reactions: Haldane relationship.

Unit 5. Inhibition of enzyme catalysis.

Inhibition of enzymatic catalysis: types of inhibitors. Reversible inhibitors: competitive inhibition, acompetitive and mixed inhibition (includes non-competitive inhibition). General model. Graphic analysis of the different types of inhibition. Determination of the inhibition constants. Concept of IC50 and its relation with the inhibition constants. Inhibition by excess substrate. Discrimination between competing substrates. Pseudo-irreversible inhibitors and irreversible inhibitors. Affinity labels. Suicide inhibitors. Use of enzyme inhibitors as drugs.

Unit 6. Analysis of enzyme kinetics in reactions with more than one substrate.

Reactions with more than one substrate: Cleland notation. Sequential ordered mechanism, statistical sequential mechanism, double displacement mechanism (ping-pong). Mathematical treatment and graphical analysis. Methods for determining the type of mechanism. Isotopic exchange and isotopic effect.

Unit 7. Kinetics of transient states.

Characteristics of rapid kinetic methods. Mixing methods: continuous flow, stopped flow and quenched-flow. Relaxation methods: temperature jump (T-jump), pressure jump (P-jump). "Bursts" and "lags". Analysis of the "Burst" of a reaction: determination of the concentration of active centers. Application of the fast reaction kinetics to the nitrogen assimilation process.

Unit 8. Effect of pH and temperature on enzymatic reactions.

Action of the temperature on enzyme kinetics. Representation of Arrhenius. Enzymes of extremophile organisms. Effects of pH on enzyme kinetics. Ionization of essential residues. Influence of pH on the kinetic parameters. Evaluation of ionization constants. Identification of the ionizable groups involved in the processes of binding and catalysis. Effects of the micro environment on the pK. Exemples.

Unit 9. Cooperativity and Allosterism.

Ligand binding to proteins. Concept and types of cooperativity. Analysis of cooperativity. Union of oxygen to hemoglobin. Cooperativity models. Model of Monod, Wyman and Changeux. Explanation of the homotropic cooperative effects by the MWC model. Allosteric enzymes. Example of enzyme with allosteric regulation: aspartate carbamyl transferase.

Unit 10. Enzymatic specificity.

The active center, specificity and three-dimensional structure. Definition of active center. Characteristics of the active center. Theories about the coupling between the enzyme and the substrate. Fisher's theory (key-lock). Koshland theory (induced-fit). Hexokinase as an example of induced coupling. Hypothesis of three-point union. Hypotheses involving tension. Stabilization of the transition state. Evidence supporting the theory of the transition state. Catalytic antibodies and their applications.

Unit 11. Study of the active center.

The active center. Identification of the binding and catalytic centers. Labelling with a part of the substrate. Use of artificial substrates. Chemical modification with specific irreversible inhibitors. Affinity labels. Suicide inhibitors, examples with pharmacological interest. Directed mutagenesis. Serine proteases: subtilisin. Comparison of mutagenesis and chemical labeling. Investigation of the three-dimensional structure of proteins: X-rays, NMR, molecular modeling. Restriction endonucleases. "Editorial" and error correction mechanisms: aminoacyl-tRNA synthetases.

Unit 12. Mechanisms of enzymatic catalysis.

Mechanisms of catalysis. Introduction to the mechanisms of enzymatic action. Acid-basic catalysis. Covalent catalysis. Pyridoxal phosphate. Catalysis with metal ions. Mechanisms of alcohol dehydrogenase and carbonic anhydrase. Environmental effect: electrostatic catalysis. The lysozyme Mechanism of subtilisin. Superoxide dismutase. Effects of proximity and orientation.Channeling intermediaries. Multifunctional enzymes. Enzymeswith additional non-enzymatic functions "moonlighting enzymes".

Unit 13. Cofactors and ribozymes.

Cofactors and ribozymes. Catalytic activity of RNA. Type of ribozymes. The ribosome is a ribozyme. Biological meaning of ribozymes. Applications of ribozymes.

Unit 14. Regulation of enzymatic activity.

Regulation of enzyme activity. Modification of the enzyme concentration. Regulation of the synthesis and degradation of enzymes. Degradation mechanisms. Variation of the enzymatic speed in function of the concentration of substrate, product and cofactors. Activation by precursor and retro inhibition. Functional meaning of cooperativity and allosterism. Hormonal control. Isozymes. Polymerization-depolymerization. Binding to other proteins. Irreversible covalent modification. Reversible covalent modification. Enzymatic cascade systems.

Unit 15. Biomedical and biotechnological applications of enzymes.

Enzymes in clinical biochemistry and biotechnology. Enzymes as therapeutic agents. Enzyme indicators of pathologies. Plasma enzymes. Factors that affect the levels of plasma enzymes. Examples of enzymes with diagnostic interest. Aminotransferases. Creatine kinase. Lactate dehydrogenase. Indicators of myocardial

infarction. Enzymes as reagents in clinical biochemistry. Enzymes and inborn errors of metabolism, examples. Enzymes in the industry. Large scale production of enzymes. Applications: drugs, food industry, detergents, textile industry. Immobilized enzymes. Enzymes as biosensors.

Unit 16. Directed evolution.

Methods to improve biocatalysts. Design and synthesis of new catalysts. Directed evolution. Generation of mutants. Selection and screening of the desired enzymatic activity. Re-design of enzymes to modify their thermostability and enantioselectivity. Adaptive evolution in the laboratory.

PROBLEMS

There will be five problem solving sessions, in which problems of enzyme purification, determination of kinetic parameters in the absence and presence of inhibitors, as well as characterization of mechanisms of inhibition and elucidation of bi-substrate reaction mechanisms will be addressed.

Delivery of works through the tool of the "Virtual Campus":

Two works will be proposed through the Virtual Campus, which must be worked out by the teams (of two/three people) established at the beginning of the course. The works must be delivered before a specific date through the Virtual Campus.

PRACTICAL LABORATORY

They are organized in 2 sessions of 4 hours in the laboratory, a session of one hour at the Chemical Analysis Service and a session of three hours in a computer room.

Program: Characterization of an enzyme overexpressed in yeast (*Saccharomyces cerevisiae)*. Analysis of the stereospecificity of the reaction for different substrates using gas chromatography. Determination of kinetic parameters in steady state conditions, using specific "software". There will be also an introduction to a software to study the tridimensional structure of proteins.

Activities and Methodology

Title	Hours	ECTS	Learning Outcomes
Type: Directed			
Problems solving classes	5	0.2	KM34, SM33, KM34
Sessions of laboratory work	12	0.48	KM34, SM33, KM34
Theory classes	35	1.4	KM34, SM33, KM34
Type: Supervised			
Group tutorials	2	0.08	KM34, SM33, KM34
Type: Autonomous			
Delivery of team work through the "Campus virtual"	11	0.44	CM32, KM34, SM33, CM32
Problem resolution	20	0.8	KM34, SM33, KM34
Study	50	2	CM32, KM34, SM33, CM32

The Biocatalysis course includes theoretical sessions, group work submissions via the Virtual Campus, problem-solving classes, and laboratory practicals. Below is a description of the structure and teaching methodology that will be followed in these activities.

Theory Classes:

The theoretical content will be delivered primarily through lectures supported by audiovisual material. Presentations used in class will be made available on the course's Virtual Campus prior to the start of each topic. These lecture-based sessions will form the core of the theoretical component.

It is recommended that students have access to the materials published on the Virtual Campus in advance to better follow the sessions. To consolidate and clarify the topics covered, it is also advised to regularly consult the books listed in the Bibliography section of this guide, as well as the topic-specific resources and links, which include videos and animations related to the content presented in class.

Group Work Submission:

This activity aims to develop teamwork skills by organizing learners into groups where all members are expected to contribute actively to both the preparation and presentation of the assignments.

The methodology will be as follows:

At the beginning of the course, students will be organized into groups of two or three people and must register their group via the Virtual Campus before the deadline set by the teaching staff. Each group will work on the assigned topics outside regular class hours. Assignment instructions and deadlines will be published on the Virtual Campus. Thegrade awarded will apply to all group members.

Problem-Solving Classes (PAUL):

There will be five sessions dedicated to solving problems related to the theoretical content. These sessions aim to consolidate prior knowledge covered in lectures and to familiarize students with experimental strategies, the interpretation of scientific data, and solving problems based on real experimental cases.

Problem sets will be shared in advance through the Virtual Campus.

Laboratory Practicals (PLAB):

There will be two 4-hour sessions, one 1-hour session, and one 3-hour session covering the following:

1. Determination of Bdh1p enzyme activity in yeast extracts (with overexpression of the enzyme). Calculation of activity in U/mL using different substrates.

2. Determination of kinetic parameters for Bdh1p using acetoin as substrate. Preparation of reaction mixtures and measurement of initial velocities. Use of a spreadsheet to calculate kinetic parameters.

3. Separation and identification of substrates and products using ethyl acetate extraction and gas chromatography with a chiral column.

4. Use of computational tools to determine kinetic parameters and inhibition patterns of Bdh1p. Structural analysis of enzymes using visualization software.

Tutorials:

One group tutorial session will be scheduled prior to each of the two midterm tests. Individual tutorials will be available upon request. If the number of requests is high, additional classroom tutorials will be arranged and announced through the Virtual Campus. These sessions aim to clarify doubts, review fundamental concepts, and provide guidance on consulted sources of information.

Materials Available on the Course's Virtual Campus:

- Presentations used in theoretical sessions

- Assignment guidelines and submission portals
- Problem statements
- Practical class protocols
- Calendar of teaching activities (lectures, tutorials, and assessments)

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.

Assessment

Continous Assessment Activities

Title	Weighting	Hours	ECTS	Learning Outcomes
Elaboration of the memory of practical work in the laboratory	15	8	0.32	CM32, KM34, SM33
Problem-solving exam	10	2	0.08	CM32, SM33
Teamwork delivered through the Virtual Campus Platform	15	0	0	CM32, KM34, SM33
Theory partial examinations	60	5	0.2	KM34, SM33

The course includes two assessment modalities: continuous and single evaluation.

Continuous assessment

The aim of continuous assessment is to encourage student engagement throughout the course and allow evaluation of their progress and understanding.

Partial theory exams - Individual assessment (6/10)

- This assessment consists of two written exams in which students must demonstrate their grasp of theoretical concepts.
- Each exam accounts for 30% of the total grade. The first will be scheduled mid-semester and the second at the end. Both exams will include multiple-choice and short-answer questions related to lecture content.

Problem-solving test - Individual assessment (1/10)

On the day of the second theory exam, students will solve three problems similar to those worked on in problem-solving sessions. This test represents 10% of the total grade.

Assignments via the Virtual Campus - Group assessment (1.5/10)

Two assignments related to theoretical and problem content will be submitted during the course in groups of 2-3 people via the Virtual Campus. Evaluation will consider not only correctness but also structure and presentation. The entire group will receive the same grade. If needed, faculty may request an individual questionnaire about the group work. If a member is clearly found not to have participated, their mark may be excluded or reduced.

Attendance and lab report - Group assessment (1.5/10)

Students must bring appropriate lab materials (lab coat, goggles, and the pre-read lab guide). Both participation and performance in the lab will be assessed. A lab report answering assigned questions must be submitted on the date set by the teaching staff. Attitude counts for 25% and the written report for 75% of the final grade for this component.

Resit exam

Students who do not achieve a minimum score of 5 will be required to take a resit exam, where they can choose to be tested on theory (part 1 and/or part 2) and/or problems.

Assignments and the practical report are not resit-eligible.

Single assessment

Theory (60%)

An individual final exam held on the same day as the second theory exam, covering the full syllabus. It will include multiple-choice and short-answer questions and represent 60% of the final grade.

Problems (10%)

Held on the same day as the theory exam, this test will consist of three exercises from across the course content and count for 10% of the final grade.

Virtual Campus assignments (15%)

Content and criteria are the same as those described for continuous assessment.

Attendance and lab report (15%)

Conditions are the same as those set out under continuous assessment.

Written communication skills will also be considered in all cases.

Resit exam

Students who do not achieve a minimum grade of 5 must take the resit, choosing to be tested on theory and/or problems.

Assignments and the practical report are not resit-eligible.

Global course assessment

- Under continuous assessment, the final grade will include results from the two partial theory exams, the problem test, group assignments, and practical attendance and report. A minimum of 5 out of 10 is required to pass.
- Under single assessment, the final grade will include the final theory and problem tests, group assignments, and the practical component. A minimum of 5 out of 10 is also required to pass.

Justified absences

Students who miss an exam for a justified reason (illness, death of a first-degree relative, accident) and provide official documentation to faculty or the degree coordinator will be allowed to reschedule the exam.

Bibliography

Grunwald, Peter. *Biocatalysis: Biochemical Fundamentals and Applications / Peter Grunwald, University of Hamburg, Germany*. Second edition. New Jersey: World Scientific, 2018. Print.

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Cornish-Bowden, Athel. *Fundamentals of Enzyme Kinetics / Athel Cornish-Bowden*. 4th., completely rev. and greatly enl. ed. Weinheim: Wiley-VCH, 2012. Print.

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Web links

They will be updated in the Virtual Campus ofthe subject.

Software

Software

The software that will be used during the course will be:

EXCEL. Spreadsheet software to organize, calculate, and visualize numerical data.

GRAFIT. Software for fitting experimental data to mathematical models.

COPASI. It is a program for the simulation and analysis of biochemical and dynamic networks.

PYMOL. It is a molecular visualization program.

JSME i CHEMSKETCH. Software that allows the drawing of the structures of chemical compounds.

Groups and Languages

Please note that this information is provisional until 30 November 2025. You can check it through this link. To consult the language you will need to enter the CODE of the subject.

Name	Group	Language	Semester	Turn
(PAUL) Classroom practices	441	Spanish	second semester	morning-mixed
(PLAB) Practical laboratories	441	Spanish	second semester	afternoon
(TE) Theory	44	Spanish	second semester	morning-mixed