Universitat Autònoma de Barcelona

2023/2024

Biocatalysis

Code: 100867
ECTS Credits: 6

Degree	Type	Year	Semester
2500252 Biochemistry	OB	2	2

Contact

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Teaching groups languages

You can check it through this link. To consult the language you will need to enter the CODE of the subject.
Please note that this information is provisional until 30 November 2023.

Teachers

Mohammed Moussaoui Keribii

Prerequisites

There are no official prerequisites. In any case, some of the contents of the 1st and 2nd year subjects (first semester) are necessary to be able to follow the subject correctly. In particular, those of the following subjects: Thermodynamics and Chemical Kinetics, Organic Chemistry of Biochemical Processes, Biochemistry I, Biochemistry II, Chemistry and Protein Engineering, Basic and Advanced Instrumental Techniques.
For some activities, a basic level of reading comprehension of English is required.

Objectives and Contextualisation

The subject Biocatatalization focuses on the study of enzymes. The knowledge of enzymes is key in the Biochemistry and related sciences given their role as catalysts of biological reactions and applications in biotechnological processes. The subject analyzes enzymes from different perspectives: 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 the biotechnological and biomedical applications.
Specific objectives of the subject:
Knowledge of the general characteristics, classification and testing methods of enzymatic activity.
Analysis of enzymatic kinetics and determination and meaning of kinetic parameters.
Knowledge of enzymatic 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.

Competences

- Act with ethical responsibility and respect for fundamental rights and duties, diversity and democratic values.
- Demonstrate understanding and use of the mechanisms of biological catalysis based on the structure of biological catalysts and chemical reactions.
- Interpret experimental results and identify consistent and inconsistent elements.
- Introduce changes in the methods and processes of the field of knowledge to provide innovative responses to the needs and demands of society.
- Manage bibliographies and interpret the information in the main biological databases, and also know how to use basic ICT tools.
- Manage information and the organisation and planning of work.
- Read specialised texts both in English and one's own language.
- Take account of social, economic and environmental impacts when operating within one's own area of knowledge.
- Take sex- or gender-based inequalities into consideration when operating within one's own area of knowledge.
- Use ICT for communication, information searching, data processing and calculations.
- Use analytical methodologies for assaying the biological activity of cellular components, especially enzymes, both in vitro and in vivo.

Learning Outcomes

1. Act with ethical responsibility and respect for fundamental rights and duties, diversity and democratic values.
2. Assess the suitability of the methods for determining enzyme activities and analyse the effect of the test conditions.
3. Calculate and interpret the kinetic parameters of enzyme reactions, by means of graphic methods using computer programmes.
4. Explain the fundamental physicochemical principles of enzyme catalysis.
5. Explain the structural bases and the principal mechanisms of enzyme catalysis and how it is regulated.
6. Interpret experimental results and identify consistent and inconsistent elements.
7. Introduce changes in the methods and processes of the field of knowledge to provide innovative responses to the needs and demands of society.
8. Manage information and the organisation and planning of work.
9. Obtain information from databases on the structure, activity, and biological functions of enzymes and their applications.
10. Read specialised texts both in English and one's own language.
11. Take account of social, economic and environmental impacts when operating within one's own area of knowledge.
12. Take sex- or gender-based inequalities into consideration when operating within one's own area of knowledge.
13. Use ICT for communication, information searching, data processing and calculations.

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. K-systems and V-systems. Koshland, Nemethy and Filmer model. Determination of the cooperative model that follows a certain enzyme. 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.

The problems that are proposed refer to the analysisof enzyme activity and determination and interpretation of kinetic parameters. The statements of the problems will be delivered through the Virtual Campus.

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 three/four people) of students established at the beginning of the course. The works must be delivered before a specific date through the Virtual Campus.

Methodology

The subject of Biocatalysis consists of theoretical classes, problem solving classes and use of computer applications, resolution and delivery of group questions and tutorials. The training activities of the subject are complemented by the practical contents of training in the field of enzymes taught in the course Integrated Laboratory 4. The following describes the organization and teaching methodology that will be followed in these types of training activities.

Theory classes:

The content of the theory program will be taught mainly by the teacher in the form of master classes with audiovisual support. The presentations used in class by the teacher will be available in the Virtual Campus of the subject before the start of each of the topics of the course. These expository sessions will be the most important part of the theory section. It is recommended to have the material published in the Virtual Campus to be able to follow the classes more comfortably. In order to consolidate and clarify the contents explained in class, it is advisable to consult regularly the books recommended in the Bibliography section and the links and resources indicated in the different topics, which contain information related to the processes explained in class.

Classes of problem solving and use of computer applications:

In these sessions the class group will be divided into two groups (A and B). The students should check the group to which they belong and attend the corresponding classes. There will be 10 problem sessions that will be devoted to solving problems related to the contents of the theory program and the use of computer applications related to enzymes.

It is intended that these classes serve to consolidate the contents previously worked in the theory classes and also to know some of the experimental strategies, the interpretation of scientific data and the resolution of problems based on real experimental situations.

Resolution and delivery of teamwork:

This activity aims to work on the competence of teamwork, through the organization of students in working groups in which all members must actively participate in the resolution of problems.

The methodology of this activity will be the following:

At the beginning of the course the students will be organized in groups of four people, registering the groups through the Virtual Campus before the deadline indicated by the teacher.
The groups will work the problems indicated for this activity outside of class time.
The works will be delivered through the Virtual Campus. The qualification obtained will be applicable to all the members of the working group to which the student belongs.

The delivery statements will be published through the Virtual Campus where the delivery dates will also be indicated.

Tutorials

Individual tutorials will be carried out at the request of the students. In the event that the number of applications was extremely high, especially in the face of partial examinations, a classroom tutorial could be held before each partial of theory (two in total), that would be announced through the Virtual Campus in due time. The objective of these sessions will be to solve doubts, review basic concepts and guide on the sources of information to consult. These sessions will neither be used to expose new topics nor to advance in the theory program but they will be sessions of debate and discussion.

Material available in the Virtual Campus of the subject:

Presentations used by the teacher in theory classes.

Statements of the problems or cases to work to the classes of problems. It will include the problems of group work.

Programming and information on teaching activities (classroom classes, tutorials, evaluations, ...).

Questions towork in group teams.

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			
Problem solving classes and the use of computer applications	10	0.4	13, 2, 3, 6, 10, 9
Theory classes	35	1.4	1, 12, 11, 2, 3, 4, 5, 7, 10, 9
Type: Supervised			
Group tutorial	2	0.08	2, 3, 4, 5
Type: Autonomous			
Analysis and problem solving	20	0.8	13, 2, 3, 8, 6, 10, 9
Study	57	2.28	13, 4, 5, 8, 6, 10
Teamwork resolution of problems and delivery through the Campus Virtual Platform	19	0.76	13, 2, 3, 8, 6, 10, 9

Assessment

This subject includes two types of assessment: continuous and unique.

Continuous assessment.

The objective of continuous assessment is to encourage the student's effort throughout the course, allowing them to evaluate their degree of follow-up and understanding of the subject.

Theory (70% of the overall grade)

Individual evaluation through:

Two partial tests with multiple-choice questions and short questions, which will be eliminatory if their qualification is equal to or greater than 4 (out of 10). The weight of each test will be 35% of the overall grade.

A test of recovery of theory partials with multiple-choice questions and short questions corresponding to the first and/or second partials. To participate in the recovery, students must have been previously evaluated in a set of activities whose weight equals to a minimum of two thirds of the total grade of the subject or module.

Therefore, students will obtain the "non-Valuable" qualification when the evaluation activities carried out have a weighting of less than 67% in the final grade.

Students who have obtained a grade lower than 4.0 (out of 10) in the first and/or partial will have to take the recovery exam of the corresponding partial (s) (first partial, second partial or both).

The total weight of the theory evaluation will be 70% of the overall grade.

Problems (20% of the overall grade)

1-Individual evaluation:

Two partial tests with problems, which will be eliminatory if their qualification is equal to or greater than 4 (out of 10). The weight of each test will be 10% of the overall grade.

A test of recovery of partial problems with problems corresponding to the first and/or second partials. Students who have obtained a grade lower than 4.0 (out of 10) in the previous first and/or second partial will have to take the recovery exam of the corresponding partial (s) (first partial, second partial or both).

The weight of the individual evaluation of problems will be 20% of the overall grade.

In all cases, in addition to knowledge, the acquisition of written communication skills will be considered.

Assignments through the Virtual Campus: (10% of the overall grade)

Periodically (2 times during the course), two works will be proposed that must be solved before a specific date. The works prepared in groups of 3 or 4 people will be delivered through the Virtual Campus. For the assessment, not only the correct resolution of the work but also its approach and presentation will be considered. The whole group will receive the same grade. If deemed necessary, the teacher may request that a questionnaire regarding the group's work be completed individually. Although the results of this questionnaire will not initially have a specific weight in the grade of the subject, in case of detecting negative evaluations of a person by the rest of the members of their group that show that they have not participated in the work, the qualification obtained by the group will not be applied or may be reduced. The weight of each delivery will be 5% of the overall grade.

The total weight of the evaluation through the assignments by the Virtual Campus will be 10% of the overall grade.

Examination-based assessment.

Theory (70% of the overall grade)

Individual evaluation through:

A final test, which will be carried out simultaneously with the second partial exam of the subject, in which the questions will be of the whole matter of the subject. In this test there will be multiple choice questions and short questions. The weight of this test will be 70% of the overall grade.

Problems (20% of the overall grade).

A final test, which will be carried out simultaneously with the second partial exam of the subject, in which problems of all the matter of the subject must be solved. The weight of this test will be 20% of the overall grade.

Assignments through the Virtual Campus: (10% of the overall grade).

The content and rules of this section are the same as those described under the heading of continuous assessment.

In all cases, in addition to knowledge, the acquisition of written communication skills will be considered.

Theory and/or problems recovery test.

Students who have obtained a grade lower than 4.0 (out of 10) in the single assessment test, in the theory and / or problems part, will have to take the corresponding recovery exam: theory and / or problems.

Global evaluation of the subject.

In case of continuous evaluation, the global evaluation of the subject will include the qualifications of the two partial tests of both theory and problems, as well as the delivery of group work. Out of a total of 10 points, it will be necessary to obtain a global grade equal to or greater than 5 points to pass the subject.

In the case of single evaluation, the overall evaluation of the subject will include the qualification of the final test of theory and problems, as well as the delivery of group work. Out of a total of 10 points, it will be necessary to obtain a global grade equal to or greater than 5 points to pass the subject.

Students who cannot attend an individual assessment test for justified reasons (such as illness, death of a first-degree relative or accident) and provide the corresponding official documentation to the teacher or degree coordinator, will be entitled totake the test in question on another date.

Assessment Activities

Title	Weighting	Hours	ECTS	Learning Outcomes
Continuous evaluation: two partial theory tests. Examination-based evaluation: final theory test.	70%	4	0.16	1, 12, 11, 2, 4, 5, 8, 6, 7, 10
Evaluation of homework sent by the Virtual Campus	10%	0	0	13, 2, 4, 5, 8, 10, 9
Resolution of problems and practical cases and use of computer applications	20%	3	0.12	13, 2, 3, 8, 6, 9

Bibliography

Specific Titles

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Links

They will be updated in the Virtual Campus of the subject

Software

Software

The programs that will be used during the course are:

COPASI.

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

http://copasi.org/

PYMOL.

It is a molecular visualization program.

https://pymol.org

JSME and CHEMSKETCH.

Software that allows the drawing of the structures of chemical compounds.