



1. General information

Course: ENZIMOLOGY

Type: CORE COURSE

Degree: 341 - UNDERGRADUATE DEGREE PROGRAMME IN BIOCHEMISTRY

Center: 501 - FACULTY OF ENVIRONMENTAL SCIENCES AND BIOCHEMISTRY

Year: 2

Main language: Spanish

Use of additional languages:

Web site:

Code: 13316

ECTS credits: 6

Academic year: 2023-24

Group(s): 40

Duration: C2

Second language: English

English Friendly: Y

Bilingual: N

Lecturer: ELENA BONZÓN KULICHENKO - Group(s): 40				
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ICAM, despacho 0.30	QUÍMICA INORG., ORG., Y BIOQ.	926051477	Elena.Bonzon@uclm.es	By appointment by email
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Lecturer: MARIO GUTIÉRREZ TOVAR - Group(s): 40				
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Lecturer: ANA MARIA RODRIGUEZ CERVANTES - Group(s): 40				
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Lecturer: MARIA RODRIGUEZ PEREZ - Group(s): 40				
Building/Office	Department	Phone number	Email	Office hours
Edif. 6. Dcho 11	QUÍMICA INORG., ORG., Y BIOQ.	5435	maria.rodriguezperez@uclm.es	Mondays and Wednesdays from 12 to 14 h, Previous appointment by e-mail is required.

2. Pre-Requisites

Knowledge of Biochemistry, General Chemistry and Basic Mathematical Analysis. Teaching will be developed assuming that students have the optimal level of knowledge corresponding to the subjects of Fundamentals of Biochemistry, and Thermodynamics and Kinetics, corresponding to the first year of the degree.

3. Justification in the curriculum, relation to other subjects and to the profession

The subject is in the second year of the degree and aims to develop with greater intensity the knowledge of the functional or kinetic properties of enzymatic catalysis, acquired by students in the subjects Thermodynamics and Kinetics and Fundamentals of Biochemistry, of the first year of the Degree.

The contents of this subject are framed within the Degree in the second year, as a previous and essential training for the understanding and integration of knowledge that will be taught in subsequent courses of the degree, mainly within the subjects of Human Physiology and Metabolism and its Regulation in the third year of the Degree.

Likewise, the training received in the subject will be essential to achieve a professional competitive advantage, the mastery of enzymes as main elements or important tools, in preparative, degradative and synthetic analytical applications. This will enhance the performance of jobs in public and private companies, dedicated to health biotechnology (red), agri-food (green), industrial (white) and business management (gray), both in R+D and business management departments on knowledge, occupational risks, environment and quality.

4. Degree competences achieved in this course

Course competences

Code	Description
E01	Express themselves correctly in basic biological, physical, chemical, mathematical and computer terms.
E02	Work properly and quality driven in a chemical, biological and biochemical laboratory, including safety, handling and disposal of waste and keeping a record of activities.
E03	Understand and know how to explain the physical and chemical bases of biochemical processes and the techniques used to investigate them
E13	Correct handling of different computer tools
E15	Experimentally determine the concentrations of metabolites, the kinetic and thermodynamic parameters and the control coefficients of the reactions of the intermediate metabolism.
E21	Understand the chemical and thermodynamic principles of biocatalysis and the role of enzymes and other biocatalysts in the functioning of cells and organisms.
G01	To possess and understand the knowledge in the area of Biochemistry and Molecular Biology at a level that, based on advanced

G02	textbooks, also includes cutting-edge aspects of relevance in the discipline. To know how to apply the knowledge of Biochemistry and Molecular Biology to professional practice and to possess the necessary intellectual skills and abilities for this practice, including the capacity for: information management, analysis and synthesis, problem solving, organization and planning and generation of new ideas.
G03	Be able to collect and interpret relevant data, information and results, draw conclusions and issue reasoned reports on relevant social, scientific or ethical issues in connection with advances in Biochemistry and Molecular Biology.
G04	To know how to transmit information, ideas, problems and solutions in the field of Biochemistry and Molecular Biology to a specialized and non-specialized public.
G05	Develop those strategies and learning skills necessary to undertake further studies in the area of Biochemistry and Molecular Biology and other related areas with a high degree of autonomy.
G06	Acquire skills in the handling of computer programs including access to bibliographic, structural or any other type of databases useful in Biochemistry and Molecular Biology.
T10	Ability to self-learn and to obtain and manage bibliographic information, including Internet resources

5. Objectives or Learning Outcomes

Course learning outcomes

Description

6. Units / Contents

Unit 1: Enzymatic kinetics

- Unit 1.1 Hyperbolic kinetics.
- Unit 1.2 Enzyme-substrate complex.
- Unit 1.3 Michaelis-Menten equation. Equilibrium approximation: Henri and Michaelis-Menten model.
- Unit 1.4 Steady state approximation: Briggs and Haldane modification.
- Unit 1.5 Pre-steady state: kinetic treatment and experimental techniques.

Unit 2: Kinetic parameters

- Unit 2.1 Meaning of K_m , V_{max} and k_{cat} .
- Unit 2.2 Affinity and K_s .
- Unit 2.3 Specificity constant.
- Unit 2.4 Catalytic efficiency.
- Unit 2.5 Relationship between kinetic parameters and the equilibrium constant of the reaction: Haldane equation.

Unit 3: Experimental determination of kinetic parameters

- Unit 3.1 Linearization of the Michaelis-Menten equation: Lineweaver-Burk, Hans-Woolf, Eadie-Hofstee and Eisenthal/Cornish-Bowden graphs.
- Unit 3.2 K_m and k_{cat} in enzymatic reactions with more than one intermediate complex.

Unit 4: Enzyme inhibition

- Unit 4.1 Reversible inhibition: competitive, acompetitive and mixed inhibitions; Kinetic analysis, inhibition constants and representations
- Unit 4.2 Irreversible inhibition: covalent modification of enzymes, inhibitor efficiency; Inactivation constant. Classification of irreversible inhibitors; structural and kinetic characteristics. Applications of the study of enzyme inhibition.

Unit 5: Kinetics of multisubstrate reactions

- Unit 5.1 Bisubstrate reactions
- Unit 5.2 Reaction mechanisms
- Unit 5.3 Cleland's terminology
- Unit 5.4 Kinetics of bisubstrate reactions; Alberty's general equation.
- Unit 5.5 Determination of reaction mechanism and kinetic constants; Primary and secondary representations.
- Unit 5.6 Discrimination between types of mechanisms; Steady-state kinetic analysis: inhibition by product.

Unit 6: Effect of reaction medium on enzyme activity

- Unit 6.1 Effect of pH on reaction rate; Kinetic analysis.
- Unit 6.2 Identification of the amino acids involved in the catalytic process; determination of pK_e and pK_{es}
- Unit 6.3 Effect of temperature on reaction rate.

Unit 7: Mechanism of action of enzymes

- Unit 7.1 Transition state theory
- Unit 7.2 Activation energy
- Unit 7.3 Factors responsible for the catalytic power of enzymes.
- Unit 7.4 Intramolecular catalysis. Entropic effect
- Unit 7.5 Formation of the enzyme-substrate complex; binding energy.
- Unit 7.6 Molecular mechanisms of binding energy utilization
- Unit 7.7 Enzyme-substrate and enzyme-transition state complementarity.
- Unit 7.8 Preferential union to the transition state: induced fit model and link distortion model.

Unit 8: Mechanisms of enzymatic catalysis

- Unit 8.1 Acid-base catalysis.
- Unit 8.2 Electrostatic catalysis; Electrophilic catalysis by metal ions
- Unit 8.3 Covalent catalysis: nucleophilic and electrophilic
- Unit 8.4 Amino acids involved.
- Unit 8.5 Representative examples: protease reaction mechanisms; Imines and reaction mechanism of aldolase.
- Unit 8.6 Enzymatic catalysis involving coenzymes

Unit 9: Enzyme regulation

- Unit 9.1 Regulation of metabolic pathways
- Unit 9.2 Teoría del Análisis del Control Metabólico. Coeficiente de control de flujo.
- Unit 9.3 General and specific mechanisms of regulation of metabolic pathways. Coefficients of elasticity and response.
- Unit 9.4 Regulatory stages.
- Unit 9.5 Mechanisms of regulation of the amount of an enzyme. The proteasome.

Unit 10: Cooperativity**Unit 10.1** Ligand-protein interactions.**Unit 10.2** Cooperativity. Dynamics of oxygen-binding proteins.**Unit 10.3** Hill's equation. Determination of the Hill index: Logarithmic representation of Hill.**Unit 10.4** Scatchard's equation and its logarithmic representation.**Unit 10.5** Adair equation.**Unit 10.6** Saturation ratio (cooperativity index)**Unit 11: Allosteric effects****Unit 11.1** Sigmoid kinetics and allosteric enzymes.**Unit 11.2** Mechanisms of allosteric interactions and cooperativity**Unit 11.3** Monod, Wyman and Changeux symmetry model.**Unit 11.4** Sequential model of Koshland, Nemethy and Filmer.**Unit 12: Reversible covalent modification****Unit 12.1** Oxidative modifications.**Unit 12.2** Phosphorylation-dephosphorylation reactions.**Unit 12.3** Protein kinases and phosphoprotein phosphatases. Mechanisms of action and activation.**Unit 12.4** Other forms of enzyme regulation by covalent modification.**Unit 13: Regulation by proteolytic activation****Unit 13.1** Mechanisms of proteolytic activation.**Unit 13.2** Activation of digestive enzymes.**Unit 13.3** Proteolytic processing of procollagen.**Unit 13.4** Proteolytic processing of HIV polyprotein.**Unit 13.5** Cascade of blood coagulation.**Unit 13.6** Activation of caspases and apoptosis.**Unit 14: Organized enzyme systems. Isoenzymes.****Unit 14.1** Multienzyme proteins and multienzyme complexes.**Unit 14.2** Molecular mechanisms of substrate channeling.**Unit 14.3** Isoenzymes. General characteristics. Hexokinase as a representative example of the functional and regulatory role of isoenzymes.**Unit 15: Applied enzymology****Unit 15.1** Enzymatic biotechnology: industrial and clinical applications of enzymes.**Unit 15.2** Enzymatic diagnosis.**Unit 15.3** Enzyme therapy.**Unit 16: Laboratory practices:****Unit 16.1** Computer simulation of kinetics and enzymatic mechanisms, as well as the factors that affect them.**Unit 16.2** Kinetic study of the enzymatic activity of alpha-amylase.**Unit 16.3** Subcellular fractionation of rat liver. Obtaining the cytosolic fraction.**Unit 16.4** Determination of pyruvate kinase activity in the cytosolic fraction of rat liver, its allosteric properties and the effect of two modulators: fructose-1,6-bisphosphate and L-alanine.**7. Activities, Units/Modules and Methodology**

Training Activity	Methodology	Related Competences (only degrees before RD 822/2021)	ECTS	Hours	As	Com	Description
Class Attendance (theory) [ON-SITE]	Lectures	E01 E03 E15 E21 G05	1.16	29	N	-	Master classes in which the theoretical contents will be developed. Master classes will be available to the student in Moodle.
Laboratory practice or sessions [ON-SITE]	Practical or hands-on activities	E01 E02 E03 E13 E15 G02 G03 G04 G05 G06	0.68	17	Y	Y	Practices in the laboratory where the theoretical contents will be applied and expanded. These practices will be mandatory and will be carried out in groups. Only those students who have completed the practices can be evaluated since they will not be recoverable
Practicum and practical activities report writing or preparation [OFF-SITE]	Group Work	E01 E03 E13 E15 G03 G06 T10	0.76	19	Y	Y	Written reports or videos made by the students will be delivered, according to the indications of the responsible teacher, at the end of the practices. In case of not reaching the minimum grade (4/10) in the evaluation of the internship report, this part will be recoverable in the extraordinary or special call for completion
Problem solving and/or case studies [ON-SITE]	Problem solving and exercises	E01 E13 G02 G03 G04 T10	0.32	8	Y	N	In these classes will deepen and work on the topics exposed in the master classes through the resolution of type problems.
Study and Exam Preparation [OFF-SITE]	Self-study	G01 G03 G05 T10	2.84	71	N	-	
Final test [ON-SITE]	Assessment tests	E01 E03 E15 E21 G01 G02	0.12	3	Y	Y	The student will answer a series of questions and / or problems on the topics and practical cases developed

		G05								in the subject. The evaluation will be recoverable in the extraordinary or special call for completion.
Other on-site activities [ON-SITE]	Group Work	E01 E03 E21 G01 G03 G04 G05 T10	0.08	2	Y	N				Tutored work (in group). The student will carry out an autonomous work tutored by the teacher on a topic related to the subject.
Other on-site activities [ON-SITE]	Assessment tests	E01 E03 E15 E21 G01 G02 G05	0.04	1	Y	N				The student will solve a series of questions and / or practical problems on the topics developed in the subject.
Total:			6	150						
Total credits of in-class work: 2.4			Total class time hours: 60							
Total credits of out of class work: 3.6			Total hours of out of class work: 90							

As: Assessable training activity

Com: Training activity of compulsory overcoming (It will be essential to overcome both continuous and non-continuous assessment).

8. Evaluation criteria and Grading System			
Evaluation System	Continuous assessment	Non-continuous evaluation*	Description
Final test	70.00%	85.00%	A final test will be carried out that will take into account the theoretical knowledge of the subject, problem solving, and the knowledge acquired during laboratory practices. The final exam will correspond to 70% of the final grade. A minimum grade of 4 will be required to add the grade obtained in the final exam with the rest of the activities. The following shall also be taken into account: Correct written communication. Capacity for design, analysis and synthesis.
Practicum and practical activities reports assessment	15.00%	15.00%	The following evaluation criteria shall be taken into account: -Correction of the answers to the questions that arise about each experience. -Identification and explanation of the results. -Clarity and organization in the writing of the laboratory notebook. - Ability to work in a team.
Assessment of problem solving and/or case studies	5.00%	0.00%	In these classes we will deepen and work on the topics exposed in the master classes through the resolution of type problems. The following shall be taken into account: - Clarity in the exposure of the problem. - Correction in the resolution of problems.
Oral presentations assessment	10.00%	0.00%	Tutored work (in group): the student will carry out a work tutored by the teacher on a topic related to the subject. The following shall be taken into account: - Capacity for design, analysis and synthesis in the preparation of the works; - Correct written communication; - Ability to work in a team.
Total:	100.00%	100.00%	

According to art. 4 of the UCLM Student Evaluation Regulations, it must be provided to students who cannot regularly attend face-to-face training activities the passing of the subject, having the right (art. 12.2) to be globally graded, in 2 annual calls per subject, an ordinary and an extraordinary one (evaluating 100% of the competences).

Evaluation criteria for the final exam:

Continuous assessment:

The modality assigned by default will assume the participation of the student in the Continuous Evaluation, unless he states otherwise (before the end of the class period) by means of an email to the teacher, provided that he has not completed 50% of the evaluable activities. A minimum grade of 4 will be required to add the grade obtained in the final exam with the rest of the activities. In case of not reaching the minimum grade (4/10) in the evaluation of the internship report, the report will be recoverable in the extraordinary call. The grades obtained in the preparation of the report of practices, problem solving and group work will be taken into account both in the ordinary and in the extraordinary call. The subject will only be considered passed if the set of all evaluable activities results in a grade of 5 or higher (out of 10).

Non-continuous evaluation:

The modality assigned by default will assume the participation of the student in the Continuous Evaluation, unless he states otherwise (before the end of the class period) by means of an email to the teacher, provided that he has not completed 50% of the evaluable activities. A minimum grade of 4 will be required to add the grade obtained in the final exam (85%) with the grade of the practices (15%). In case of not reaching the minimum grade (4/10) in the evaluation of the internship report, the report will be recoverable in the extraordinary call. The grade obtained in the preparation of the practice report will be taken into account in both the ordinary and extraordinary call. The subject will only be considered passed if the set of all evaluable activities results in a grade of 5 or higher (out of 10).

Specifications for the resit/retake exam:

The grades obtained in the preparation of the report of practices, problem solving and group work will be taken into account both in the ordinary and in the extraordinary call. In case of not having reached in the ordinary call the minimum qualification in the laboratory practices, in the final test of the extraordinary call a series of questions will be included to evaluate if the student has acquired the competences related to these activities.

Specifications for the second resit / retake exam:

To pass this call there will only be a final test that will represent 100% of the grade, as long as the laboratory practices have been carried out and passed with a grade equal to or greater than 5.

9. Assignments, course calendar and important dates

Not related to the syllabus/contents

Hours hours

10. Bibliography and Sources

Author(s)	Title/Link	Publishing house	City	ISBN	Year	Description
Athel Cornish-Bowden	Fundamentals of Enzyme Kinetics	Wiley-Blackwell		978-3-527-33074-4	2012	
COPELAND R A	Enzymes: a practical introduction to structure, mechanism, and data analysis (2e)	Wiley-VCH, New York		0-471-35929-7	2000	
Cox M.M. y Nelson D.L.	Lehninger: Principios de Bioquímica (6e)	Omega		978-84-282-1486-5	2014	
Fersh A.	Structure and mechanism in protein science (2e)	W.H. Freeman and Co.			1999	
Garret R.H. & Grisham G.M	Biochemistry (5e)	Cengage Learning			2012	
Mathews CK y Van Holde KE	Bioquímica	Pearson/Addison Wesley, D.L.		84-7829-053-2	2004	
Muller-Esterl W.	Bioquímica (2e)	Reverté			2008	
NUÑEZ DE CASTRO I.	Enzimología	Pirámide	Madrid	84-368-1468-1	2001	
Paul F. Cook, W.W. Cleland	Enzyme kinetics and mechanism	New York : Taylor & Francis Group		978-0-8153-4140-6	2007	
Petsko G. y Ringe D.	Protein Structure and Function	New Science Press		0878936637	2004	
Stryer L., Berg J.M., Tymoczko J.L.	Bioquímica (7e)	Editorial Reverté S.A			2013	
Voet D., Voet J.G	Bioquímica (4e)	Editorial Médica Panamericana		950-06-2301-3	2016	
						Nomenclatura de enzimas. Normas de la IUPAC/IUBMB sobre nomenclatura de enzimas. Permite búsquedas de enzimas concretas
	http://www.chem.qmul.ac.uk/iubmb/enzyme/					
						Sociedad Española de Bioquímica y Biología Molecular - SEBBM
	http://www.sebbm.es/					
						Terminología Química y Bioquímica. Recomendaciones de la IUPAC/IUBMB sobre terminología Química y Bioquímica
	http://www.chem.qmul.ac.uk/iupac/					