

**COURSE DATA****Data Subject**

<b>Code</b>	33199
<b>Name</b>	Protein technology
<b>Cycle</b>	Grade
<b>ECTS Credits</b>	4.5
<b>Academic year</b>	2022 - 2023

**Study (s)**

<b>Degree</b>	<b>Center</b>	<b>Acad. year</b>	<b>Period</b>
1102 - Degree in Biotechnology	Faculty of Biological Sciences	4	Second term

**Subject-matter**

<b>Degree</b>	<b>Subject-matter</b>	<b>Character</b>
1102 - Degree in Biotechnology	109 - Protein technology	Optional

**Coordination**

<b>Name</b>	<b>Department</b>
MINGARRO MUÑOZ, ISMAEL	30 - Biochemistry and Molecular Biology

**SUMMARY**

Proteins play crucial roles in almost all biological processes in catalysis, signaling, and structural support. This wide variety of functions stems from the existence of thousands of proteins, each with a distinct three-dimensional structure, which enables them to interact with one or more molecules within a wide range. One of the main objectives of the present biochemistry is to determine how the amino acid sequences specified conformations, and thus the functions of proteins. Only a detailed understanding of this dual structure / function we could raise new biotechnology approaches rationally.

Often the first step in these studies is the purification of the protein of interest, either for structural studies, functional and biotechnological application. Proteins can be separated from each other based on their solubility, weight, load, and binding capacity, among other characteristics. Once a protein has been purified, functional studies can be initiated or biotechnological improvements. Automatic sequencing of peptides and the application of recombinant DNA methods are providing a considerable amount of amino acid sequence data that is opening new perspectives. Many protein sequences, often deduced from genome sequences are now available in large databases of sequences and start to predict its structure and even function.

The exploration of proteins with a wide range of physical and chemical techniques currently available has enriched our degree of knowledge of the molecular basis of life. These techniques make it possible to tackle some of the most complex in terms of molecular biology, which will undoubtedly result in



improvements in a number of biotechnological processes.

## PREVIOUS KNOWLEDGE

### Relationship to other subjects of the same degree

There are no specified enrollment restrictions with other subjects of the curriculum.

### Other requirements

The student must know the structure of the main biological macromolecules, and the forces that stabilize and allow specific interactions with other molecules. Also, students must understand the mechanisms of enzymatic reactions, kinetics and regulation.

## COMPETENCES (RD 1393/2007) // LEARNING OUTCOMES (RD 822/2021)

### LEARNING OUTCOMES (RD 1393/2007) // NO CONTENT (RD 822/2021)

- Consolidate knowledge on the structure and function of proteins acquired over the previous courses.
- Acquire a global view of cellular function where it is absolutely clear what role developing major proteins in all cell types.
- Exercise the comparative method in biotechnology.
- Acquire a phylogenetic and evolutionary vision of proteins.
- Familiarization with the current literature and databases related sequences and protein structures.
- Exercise the ability to synthesize scientific information.
- Exercise the applicability to a particular case of concepts, theories and general models

## DESCRIPTION OF CONTENTS

### 1. Protein structure and purification

Structural principles of proteins

Description and chemical constitution. Physical interactions that determine the properties of proteins. Amino acid hydrophobicity scales. Structural motifs: helix-alpha, beta-sheets. Domains. Structural motifs in membrane proteins.

Protein Folding

Concept. Folding in vitro. Folding mechanisms. Folding in vivo. Chaperones. Operating mechanism GroEL / GroES. Folding of secreted proteins and membrane proteins. Prediction of the structures.

Pathologies related protein malplegamiento

General concept of amyloidosis. Alzheimer's. Parkinson. Spongiform encephalopathies. Other neurodegenerative diseases.

Proteins and enzymes of biotechnological interest



Historical perspective. Choice of biocatalyst. Kinetic properties and design of a bioreactor. Growth of the biotechnology industry. Economic considerations.

Extraction, purification and protein stability.

Properties of the proteins used in its purification. Methods of extraction and separation. Fusion proteins: large-scale purification. Denaturation mechanisms.

Recombinant proteins

Reasons for the production of recombinant proteins. Cloning strategies. Cloning vectors and expression systems in prokaryotes and eukaryotes. Protein renaturation.

## **2. Biotechnological applications**

Enzyme immobilization

Definition and classification of immobilized enzymes. Methods of immobilization. Immobilization of cofactors. Characteristics of the immobilized enzymes. Applications in industry, medicine and research.

Biosensors

Biosensor concept and its historical evolution. Types of biosensors. The bioactive component: enzymes and antibodies. Transducer types: electrochemical, optical and piezoelectric. Micro-and nanoscale. Examples of biosensors for industrial application.

Catalytic antibodies

Introduction: design and generation of catalytic antibodies. Versus abzymes enzymes. Examples of reactions available. Structural information applied to the understanding of the mechanisms of catalysis. Future of catalytic antibodies.

Enzymology in non-aqueous

Introduction: cosolvents, two-phase mixtures, reverse micelles, organic solvents. Advantages of using enzymes in nonaqueous media. Basic rules for the use of enzymes in nonaqueous media. Effect of solvent on the kinetic parameters. Strategies for the increased enzyme activity in these media.

Strategies for modifying peptides and proteins

Classical chemical modification. Affinity and photoaffinity labels. Enzymatic modification using transglutaminase and glycosyltransferases. The GFP: properties and applications to the study of protein-protein interactions (FRET R BiFC).

Encapsulation and controlled release of polypeptide drugs

Design and development of strategies for the formulation and administration of peptides and proteins. Analytical methods for the assessment of encapsulated proteins. Administration using polymeric microspheres and liposomes. Development of systems and / or vectors for targeted delivery. Cell implants.



### 3. Protein design

Directed molecular evolution

Methods for generating random diversity. Genetic selection and visual tracking. Evolution of thermostable enzymes. Evolution of enzymes for use in artificial environments. Evolution of specificity and enantioselectivity. Production of enzymes for gene therapy.

Peptidotecas combinations: chemical and biological

Combinatorial library concept. Positional tracking. Methods of preparing synthetic peptidotecas. Conformational peptidoteca concept. Peptidotecas made in phage ("phage display"): types, construction, features, applications and perspectives.

### 4. Practical sessions

Molecular imprinting of lipolytic enzymes based on interfacial activation protocols.

It consists in the entrapment of lipases active conformations for subsequent use in nonaqueous media, in order to increase their catalytic efficiency in these environments of special interest for biotechnological applications.

Helix-helix packing.

Overexpression is performed heterologous column purification Ni<sup>2+</sup>-agarose electrophoretic analysis of proteins capable of dimerization. The model system used will allow the study of interactions between transmembrane helices and an experimental approach to the structural study of membrane protein folding.

## WORKLOAD

ACTIVITY	Hours	% To be attended
Theory classes	33,00	100
Laboratory practices	12,00	100
<b>TOTAL</b>	<b>45,00</b>	

## TEACHING METHODOLOGY

Lectures: conventional classroom presentation of agenda items for 26 h. Eventually, some specific aspect of the agenda may be exposed by an invited specialist. Similarly, it will endeavor to attend research seminars related to the world of proteins that can provide during term time in research centers near the University.

Practical sessions are compulsory attendance. Consist of carrying on a teaching laboratory practical sessions described above for 12 h. Students will complete the proposed experiments working in pairs. At the end of practice students must submit a practice report presenting the experimental results while the results discussed in the context of the structure and function of proteins from a probiotic.



Seminars: Students exposed in public, a research article directly related to course content or any biotechnological innovation in the use of proteins. All students are required to prepare a brief summary of all items subject to the seminars.

## EVALUATION

The quarterly nature of the subject precludes the possibility of mid-term examinations. The assessment of theoretical knowledge (8 points) will be done through a written examination which will include questions on practical sessions (2 points).

It will also assess the quality of oral presentation, participation in both the classes taught as seminars for students and written summaries of the articles used in the workshops and the reports of practices to modulate the final .

## REFERENCES

### Basic

- BRANDEN, C. & TOOZE, J. (1999). Introduction to Protein Structure. 2<sup>a</sup> ed. Garland Publishing, Inc. New York.
- BURDEN, D.W. & WHITNEY, D.B. (1995). Biotechnology: Proteins to PCR. A course in strategies and lab techniques. Birkhäuser, Boston.
- CREIGHTON, T.E. (2010). The biophysical chemistry of nucleic acids & proteins. Helvetian Press.
- FABER, K. (1997). Biotransformations in organic chemistry: a textbook. Springer-Verlag, Berlin.
- FERSHT, A.R. (1999) Structure and Mechanism in Protein Science. Freeman, New York.
- GACESA, P. & HUBBLE, J. (1987). Enzyme Technology. Taylor & Francis, New York.
- GÓMEZ-MORENO, C. & SANCHO, J. (2003). Estructura de Proteínas. Ariel Ciencia.
- HECHT, S.M. (1998). Bioorganic chemistry. Peptides and Proteins. Oxford University Press, New York.
- KELLY, J.W. & BALDWIN, T.O. (1991). Application of enzyme biotechnology. Plenum Press, New York.
- LESK, A.M. (2001). Introduction to Protein Architecture. Oxford University Press.
- LESK, A.M. (2004). Introduction to Protein Science. Oxford University Press.
- PETSKO, G.A. & RINGE, D. (2004). Protein Structure and Function. New Science Press Ltd.





RAMIREZ-ALVARADO, M. et al. (2010). Protein misfolding diseases. John Wiley & Sons Inc.

REES, A.R., et al. (1992). Protein engineering. A practical approach. IRL Press, Oxford.

WILLIAMSON, M. (2012). How proteins work. Garland Science, Taylor & Francis Group, LLC.

