UNIVERSITÀ DI PAVIA

Anno Accademico 2020/2021

INTEGRATED LABORATORY OF PHARMACEUTICAL BIOTECHNOLOGIES

Enrollment year	2018/2019
Academic year	2020/2021
Regulations	DM270
Academic discipline	BIO/13 (APPLIED BIOLOGY)
Department	DEPARTMENT OF BIOLOGY AND BIOTECHNOLOGY "LAZZARO SPALLANZANI"
Course	BIOTECHNOLOGY
Curriculum	Chem- Pharma-Tech
Year of study	3°
Period	Annual (05/10/2020 - 14/06/2021)
ECTS	12
Lesson hours	144 lesson hours
Language	Italian
Activity type	ORAL TEST
Teacher	UBIALI DANIELA (titolare) - 3 ECTS BALESTRA BARBARA - 3 ECTS CHIESA ENRICA - 0 ECTS DELL'ACQUA SIMONE - 3 ECTS DORATI ROSSELLA - 2 ECTS PERTEGHELLA SARA - 1 ECTS ROBESCU MARINA SIMONA - 0 ECTS
Prerequisites	Basic knowledge of chemistry acquired in the courses of General and Inorganic Chemistry and Laboratory, Organic Chemistry and Laboratory, and Biochemistry of the first/second year of the three-year degrees in Chemistry, Biotechnology and Biological Sciences.
Learning outcomes	The course, through an integrated multidisciplinary approach, aims at training students in: -using enzymes as biocatalysts to obtain Active Pharmaceutical Ingredients (APIs) and/or their intermediates;

-the vehiculation of API in micro- and nanocarriers;
-understanding the reactivity and structure of metalloproteins and metalloenzymes of biological interest;
-the evaluation of the effect of a drug and the analysis of genetic variants that influence the effect of the drug itself.

Furthermore, the course aims at:
-training students about the safety standards of chemical-biological laboratories;
-providing students with the method for experimental data recording, assisting them in the critical review of the results achieved as well as in the final data reporting;
-training students about the organization and attitude toward both independent and team working.

At the end of the training, students are expected to be able to critically understand and apply thereof experimental protocols in order to:
-carry out a simple enzymatic reaction (from the reaction set-up to the product downstream) and immobilize an enzyme on a solid support;
-prepare micro- and nano- biodegradable drug delivery systems, both polymeric and lipidic; furthermore, students are expected to be able characterize the systems by using physico-chemical techniques (e.g. size distribution analysis by dynamic light scattering, evaluation of surface charge, encapsulation efficiency and in vitro release studies);
-carry out enzymatic assays and enzyme kinetics studies, in particular for the study of electron transfer reactions in biological systems, by using UV-visible spectroscopic techniques, NMR (Nuclear Magnetic Resonance) and CD (Circular Dichroism);
-assess the effect of a drug by constructing a dose/response curve, estimate the protein content of a biological sample, perform a PCR (Polymerase Chain Reaction), an analysis of the restriction sites, and an electrophoretic analysis.

As a result, students will be able to generate and then process experimental data, also by using statistical and graphical tools, analyze the results in the light of the scientific literature in the field and communicate them (data reporting).

Course contents

The course is aimed at training students in:
-carrying out the process of a biocatalyzed synthesis as a whole: determination of the specific activity of the enzyme; immobilization of the enzyme on a solid support and monitoring of the immobilization process by the activity assay and protein assay (Bradford assay); determination of the activity of the immobilized biocatalyst, immobilization yield (immobilized protein and activity recovery); set-up of the enzymatic reaction, monitoring of the biotransformation (TLC, Thin Layer Chromatography), product downstream (work-up) and purification by flash chromatography; final reaction yield. Lab training includes the use of separative techniques other than chromatography (solvent extraction, filtration, distillation under reduced pressure);
-the preparation of micro and nano-size systems for the delivery of bioactive molecules and, in particular: calcium alginate-based microparticles, synthetic polymer-based nanoparticles, liposomes.

	Micro- and nano- drug delivery systems will be characterized in terms of encapsulation efficiency, size distribution and surface charge; -kinetics of peroxidase-catalyzed oxidation reactions and study of enzyme inhibition; use of NMR techniques to characterize substrates and products; use of computational algorithms for the simulation of protein-enzyme electron-transfer complexes (docking); acid-base titration and CD spectroscopy of electron transfer proteins; -constructing a dose/response curve (without the use of animals for ethical issues) for the study of the drug-receptor interaction (agonists and antagonists) and critical analysis of the obtained experimental data (graphical representation and statistical analysis); estimation of the total proteins of a sample by a colorimetric assay (Lowry assay); molecular biology techniques applied to pharmacology (pharmacogenetics): analysis of a common genetic variant that does not involve any pathological phenotype by using DNA contained in saliva, amplification of a portion of the selected gene by PCR, detection of polymorphism by digestion of amplified DNA fragments with restriction enzyme (RFLP analysis, Restriction Fragment Length Polymorphism) and visualization of the results by agarose gel electrophoresis.
Teaching methods	The course consists of lab training in Biocatalysis, Pharmaceutical Technology, Bioinorganic Chemistry, Molecular and Cellular Pharmacology; all the activities are closely interconnected. A brief presentation will be given before starting the daily scheduled activity. Students are required to record the experimental activities carried out during the 4 modules of the Integrated Laboratory of Pharmaceutical Biotechnologies in a single personal lab notebook, according to the template provided. Students will use the data recorded for the preparation of the final report, under the supervision of professors. Lab attendance is mandatory (roll call).
Reccomended or required readings	Notes and slides (the latter material will be made available in Kiro website). K. Faber. “Biotransformations in Organic Chemistry – A textbook” Springer Ed. P. Colombo et al. “Principi di Tecnologie Farmaceutiche”. Casa Editrice Ambrosiana, Milano. A.T. Florence et al. “Physical Pharmacy”. Pharmaceutical Press, London. M.E. Aulton “Pharmaceutics: the Science of Dosage Form Design”. Churchil Livingstone, New York. Rang & Dale "Farmacologia". 8° edizione, Edra Masson.
Assessment methods	Final exam: a positive evaluation of the laboratory notebook and the final report on the experimental activity carried out in the 4 modules of the course is required to pass the exam. The final report, drawn up individually or as a team, depending on the number of students of the class, must be prepared by using a specific template, which will be provided to the students at the beginning of the lab training. In the event of one or more modules have been failed, the student is required to retake the exam.

Further information

	None.
Sustainable development	

goals - Agenda 2030	$lbl_legenda_sviluppo_sostenibile