# BIRLA INSTITUTE OF TECHNOLOGY & SCIENCE, PILANI, HYDERABAD CAMPUS INSTRUCTION DIVISION, FIRST SEMESTER 2021-2022 <u>Course Handout (Part-II)</u>

03/08/2021

In addition to part I (General Handout for all courses appended to the time table this portion gives further specific details regarding the course.

Course No. : BIO G651

Course Title : PROTEIN AND ENZYME ENGINERING

Instructor-in-Charge : RAMAKRISHNA VADREVU

Team of Instructors I Shivakumar, Deepika K, Kodam Pradeep

### 1. Course Description:

Sources, isolation, purification and storage of protein and/or enzymes; kinetics of enzyme catalyzed reactions; biocatalyst reaction engineering; techniques of production and recovery of enzymes; protein and enzyme modification; clinical and industrial applications of free and immobilized enzymes

The course will provide fundamental insights into the aspects of protein engineering principles, techniques and recent advances in manipulating proteins and enzymes, and their clinical and biotechnology applications. Enzyme production, kinetics and immobilization methods will be emphasized together with case studies to reinforce the impact of protein and enzyme engineering in basic biological research and biotechnology applications. The main objective of the course is to understand the principles and procedures to altering proteins and enzymes for applications in diverse areas.

**2.Text Book (T):** Enzyme Technology, Noorlabettu Krishna Prasad, PHI Learning Private Ltd. New Delhi, 2011.

#### 3. Reference Books

**R1**. Sheldon Park and Jennifer Cochran. Protein Engineering and Design. CRC Press, 2010 **R2**. Carl Branden & John Tooze, Introduction to Protein Structure, Second Edition, Garland Publishing Inc.

In addition to the text and reference books, necessary and relevant material indicated as Articles Review(s) and will be followed.

**R3.** Protein analysis and purification: benchtop techniques/ Ian M Rosenberg (2nd Ed) Boston Birkhauser 2005.

#### 4 Course Plan:

Lec. No.	Topic	Learning Objective	Chapter in
			Text Book (Ref book)
1	Protein engineering	Perspective	Class Notes
2-11	Protein structure	Characteristics of individual amino acids and their	R2 (2-6) Class
	/function & Protein	effect on the solubility, structure and function	Notes/Published
	Folding/stability	Protein Structure Hierarchy, Classes	Research
		Protein folds and concept of active site structures	Articles
		in some protein folds. Protein structure, stability	
		and folding, CD/FL spectrophotometric methods	
		for structure and stability analysis	
11-14	Protein/enzymes	Sources	R3 (5,10)
	production	Isolation	Class Notes
		Purification methods	
		Storage of proteins	
15-23	Overview of methods in	Phage Display Systems	R1 (1-4)
	Protein Engineering	Cell Surface Display Systems	Class
		Cell Free Display Systems	Notes/Published
		Library construction	Research
			Articles

24-31	Strategies of Protein/Enzyme Engineering	Rational Methods De novo design Directed Evolution Knowledge-Based Protein Design	R1 (4,11) Class Notes Research Articles/Review S
31-33	Enzymes and Protein Modifications  Enzyme Kinetics	Properties of Enzymes Protein modifications (Mass spectrometry, Basic theory and applications in detection of proteins and protein modifications/proteomics)  Free energy of activation Factors affecting Enzyme kinetics Michaelis-Menten Theory of Enzyme kinetics Linear equations	T1 (1) Class Notes/Literature articles/reviews T1 (2) Class notes
		Enzyme inhibition	
37-40	Enzyme Immobilization	Immobilization techniques, experimental procedures, properties of immobilized enzymes, enzyme stabilization	T1 (6), Class notes, Review articles

## LIST OF EXPERIMENTS

Practical No.	Experiment Title			
1-3	Exploring primary and secondary databases on protein sequence and structural properties (EXPASY, PDB, PDBSUM) / Protein Visualization (Pymol) structure prediction software			
4	Measurement of protein secondary structure by Circular Dichroism and estimation of secondary structure content using prediction algorithms			
5-6	Measurement of fluorescence excitation and emission spectra of proteins and determination of thermal stability of proteins (protein unfolding and obtaining thermodynamic stability measurements from either CD/or FL spectroscopy)			
6-7	Protein expression in bacteria by IPTG induction (bacterial growth and concentration of IPTG optimization): SDS Page electrophoresis			
8-11	Protein Purification using AKTA FPLC (Bacterial protein expression, centrifugation, sonication, dialysis, column chromatography)			
12	ESI-MS spectrum of purified protein. Determination of molecular weight of protein(s)			

## 5. Evaluation Scheme:

Evaluation Component	Duration	Weightage	Date & Time	Remarks
		(%)		
Mid Semester Exam	90 min	25		Closed Book
Assignments and or Quizzes		20		Open Book
(Class/Take Home) Research				
Design/Presentation etc.				
	Stipulated	20		Open Book
Practical /Record Book/Class	hours in			
Interaction/viva/quiz)	the TT			
Comprehensive Exam	2 hours	35		Closed Book

**6.Chamber Consultation Hour:** Will be announced in the Class.

7.Notices: Notice, if any, concerning the course will be displayed only on CMS

**8.Make-up Policy:** For genuine cases only for Tests & Comprehensive exam. Prior permission of the Instructor-in-Charge is necessary. <u>No make-up will be granted for other components</u>. <u>No make-up for lab experiments/components</u>.

**9.Academic Honesty and Integrity Policy**: Academic honesty and integrity are to be maintained by all the students throughout the semester and no type of academic dishonesty is acceptable.

**INSTRUCTOR-IN-CHARGE**