B. Sc. BIOCHEMISTRY- III SEMESTER
SYLLABUS- THEORY AND PRACTICALS

COURSE CODE: DSC-3T: SEPBC-301
COURSE TITLE: BIOCHEMICAL TECHNIQUES AND ENZYMOLOGY (Theory)

Course title	BIOCHEMICAL TECHNIQUES & ENZYMOLOGY
Course code	DSC-3T: SEPBC-301
Course credits	03
Total contact hours	45
Duration of ESA (Hour)	03
Formative assessment marks	20
Summative assessment marks	80

Unit 1 **15 Hrs**

Biochemical Techniques**:** Chromatography, principle, procedures and applications of paper chromatography - ascending, descending, circular and 2D chromatographies.

Column chromatography- gel-filtration, adsorption- ion-exchange, and affinity chromatographies. Thin layer and hydrophobic chromatographies.

Principle and applications of HPLC, RP-HPLC and GLC.

Centrifugation: Definition, sedimentation coefficient and Svedberg's unit. Principle and types of centrifugations. Procedure of differential and density gradient centrifugations (Rate zonal and Isopycnic). Ultra centrifugation- construction and applications.

Electrophoresis: Principle, procedures and applications of paper and gel electrophoresis (Agarose, native-PAGE, SDS–PAGE). Visualizing techniques **(E**thidium bromide, Coomassie blue, and silver stain). Isoelectric focusing, Zymogram.

Unit 2 **15 Hrs**

Introduction to Enzymes: Definition, general characteristics, holoenzyme, apoenzyme, co-factors, co-enzymes, metalloenzymes, abzymes and ribozyme.

Classification of enzymes based on IUBMB with examples.

Units of enzyme activity, specific activity, enzyme specificity, concept of active site.

Theories of enzyme catalysis – Lock and key model, Koshland's induced fit theory.

Allosteric enzymes and their characteristics with PFK as an example.

Isoenzymes – Properties and significance, LDH as an example.

Multienzyme complexes - Properties and significance. Pyruvate dehydrogenase complex-subunit composition & biological role.

Applications of enzymes: Industrial & Medical applications.

Unit 3 **15 Hrs**

Enzyme kinetics & Enzyme inhibition: Enzyme Kinetics: Factors affecting rate of enzyme catalyzed reactions**.** Effect of substrate concentration, pH, temperature. Michaelis-Menten equation (derivation not required). Lineweaver-Burk (L-B) plot. Definition and significance of Km & Vmax. Turn over number.

Enzyme inhibition: Reversible enzyme inhibition- competitive, non-competitive and uncompetitive inhibitions with suitable examples. Graphical representations, effect of inhibitor on Km & Vmax using Michaelis-Menton, and LB plots. Irreversible inhibition-suicide inhibition.

REFERENCES

1. Wilson K, Walker J (2010). Principles and Techniques of Biochemistry and Molecular Biology.
2. Upadhyaya K, Upadhyaya N. Biochemistry
3. Palmer, T., & Bonner, P. L. (2004). *Enzymes: Biochemistry, Biotechnology, Clinical Chemistry* (5th ed.). Horwood Publishing.
4. Price, N. C., & Stevens, L. (1999). *Fundamentals of Enzymology: The Cell and Molecular Biology of Catalytic Proteins* (3rd ed.). Oxford University Press.

COURSE CODE: DSC-3P: SEPBC-302

COURSE TITLE: BIOCHEMICAL TECHNIQUES – PRACTICALS-3

Course title	BIOCHEMICAL TECHNIQUES PRACTICALS-3
Couse code	DSC-3P: SEPBC-302
Course credits	02
Total contact hours	4 h/week
Duration of ESA (Hour)	3
Formative assessment marks	10
Summative assessment marks	40

1. Identification of amino acids by circular paper chromatography.
2. Ascending paper chromatography of amino acids.
3. Identification and resolution of pigments by thin layer chromatography.
4. Two-dimensional chromatography of amino acids.
5. Gel-filtration chromatography-determination of void volume.
6. Column chromatography of plant pigments.
7. Electrophoretic separation of plasma proteins using polyacrylamide gel electrophoresis.
8. Agarose gel electrophoresis of DNA.
9. Separation of human lymphocytes by centrifugation.
10. Determination of packed cell volume/ hematocrit.
11. Recording the absorption spectrum of riboflavin.

B. Sc. BIOCHEMISTRY - III SEMESTER
ELECTIVE -1

COURSE CODE:
COURSE TITLE: GENETIC ENGINEERING

Course title	GENETIC ENGINEERING
Couse code	
Course credits	03
Total contact hours	45
Duration of ESA (Hour)	03
Formative assessment marks	20
Summative assessment marks	80

Unit 1 **15 Hrs**

Introduction to Genetic Engineering: Restriction and modification systems; production of DNA fragments, restriction endonucleases and other enzymes used in manipulating DNA molecules (DNA polymerases, RNA Polymerases, reverse transcriptases, ligases, taq polymerase, kinases). Ligation of DNA molecules. DNA ligase, sticky ends, blunt ends, linkers and adapters.

Plasmids: Plasmids and bacteriophages as vectors for gene cloning. Cloning vectors based on *E. coli* plasmids, pBR322, and pUC18. Cloning vectors based on M13 and λ bacteriophage. Selection of plasmids: Direct selection, marker rescue. Gene libraries, identification of a clone from gene library, colony and plaque hybridization probing, methods based on detection of the translation product of the cloned gene.

Unit 2 **15 Hrs**

Introduction of DNA into cells and selection of recombinant clones: Uptake of DNA by competent bacterial cells. Selection of transformed cells. Identification of recombinants. Sequence dependent and independent screening, southern-western, colony and plaque hybridization - insertional inactivation, blue-white colony selection. Introduction of phage DNA into bacterial cells. Identification of recombinant phages.

Expression of cloned genes: Vectors for expression of foreign genes in *E. coli*, cassettes and gene fusions.

Unit 3 **15 Hrs**

Polymerase chain reaction (PCR): designing primers for PCR. Studying PCR products. Cloning PCR products and RT-PCR. DNA sequencing: DNA sequencing by Sanger's method, modifications based on Sanger's method. Automated DNA sequencing.

Applications of genetic engineering: Applications in medicine, production of recombinant pharmaceuticals such as insulin. Recombinant vaccines. Gene therapy. Transgenic animals. Applications in agriculture - plant genetic engineering, insect resistant crops, problems with genetically modified plants, safety and ethical concerns.

Reference

1. N. Arumugam, L.M. Narayanan, A Mani, A.M Selvaraj, Padmalata Singh. Genetic Engineering, Saras Publication.
2. T. A. Brown. Gene Cloning & DNA Analysis, An introduction. (2001).

B. Sc. BIOCHEMISTRY- IV SEMESTER
ELECTIVE-1

COURSE CODE:
COURSE TITLE: IMMUNOLOGY

Course title	IMMUNOLOGY
Couse code	
Course credits	03
Total contact hours	45
Duration of ESA (Hour)	03
Formative assessment marks	20
Summative assessment marks	80

Unit 1 **15 Hrs**

Introduction: Historical background of immunology, definition, scope and significance of immunology. Important organs and cells: bone marrow, lymphnodes, thymus, spleen, white blood cells, and lymphocytes. Formation and functions of neutrophils, macrophages, T & B lymphocytes, helper T-cells and killer T-cells.

Types of immunity: classification, innate and adaptive immunity, cells and molecules involved in innate and adaptive immunity.

Unit 2 **15 Hrs**

Antigens: definition, antigenicity, haptens, epitopes, chemical nature of antigens.

Antibodies: definition, types and their functions. Structure of a typical Immunoglobulin (lgG - Light chain, heavy chain, hyper- variable regions, constant domains, Fab and Fc regions).

Antigen- Antibody interactions, cytokines- properties and functions.

Immunological techniques: precipitation reaction, immunodiffusion, RIA & ELISA

Unit 3 **15 Hrs**

Major histocompatibility complex, structure and functions of MHC molecules, endogenous and exogenous pathways of antigen processing and presentation, complement system- components and pathways of compliment activation. Immunological disorders- allergy (Types of hyper sensitivity reactions), AIDS, SCID. Vaccination - Vaccines and their preparations, adjuvants, primary and secondary immune responses.

REFERENCES

1. Owen, Judith A., Jenni Punt, and Sharon A. Stranford. Kuby immunology. New York: WH Freeman, 2013.

2. Delves, Peter J., Seamus J. Martin, Dennis R. Burton, and Ivan M. Roitt. & Roitt' essential immunology. Vol. 20. John Wiley & Sons, 2011.