

**M.Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**BC 1.1: CHEMISTRY OF BIOMOLECULES**

**Course Outcomes:**

- CO1: To offer detailed knowledge of biomolecules for living systems
- CO2: To provide basic concepts of structural organization and characterization of proteins
- CO3: To learn about Oligosaccharides and lectin interactions in biochemical processes
- CO4: To acquire knowledge on physicochemical properties and characterization of fats and oils
- CO5: To understand the structure of DNA and RNA and their types

**Course Specific Outcomes:**

- CSO1: To provide the concept of Oligosaccharides and lectin interactions in biochemical process
- CSO2: To study salient features of Bacterial and Plant lipids
- CSO3: To gain knowledge on nucleotides as energy carriers and other important functions

**Course Learning Outcomes:**

- LO1: Students will acquire an insight into various biomolecules which constitute the living organisms
- LO2: Students will learn the structure and properties of carbohydrates, proteins, lipids, cholesterol, DNA, RNA, glycoproteins, glycolipids and their importance in biological systems
- LO3: Students will develop perception on the sequencing of proteins and nucleic acids
- LO4: Students will gain knowledge on the Structure and properties of Porphyrins

**Unit – 1**

Carbohydrates: Classification, Physicochemical properties; Chemistry, Biological roles and Structural elucidation of polysaccharides - homo and heteropolysaccharides, Peptidoglycans, Glycosaminoglycans; Glycoconjugates – Proteoglycans, Glycoproteins and Glycolipids; Oligosaccharides - Lectin interactions in biochemical processes

**Unit – 2**

Amino acids: Classification, Structure and Physicochemical properties; Peptide bond, Peptides of biological importance; Chemical synthesis of peptides – Solid phase peptide synthesis; Proteins – Classification, Isolation, Purification and Characterization of proteins, Criteria of homogeneity; Protein sequencing; Structural organization of Proteins – Ramachandran plots; Denaturation of proteins

### **Unit – 3**

Lipids: Classification; Structure, Properties and Biological roles of Phospholipids and Sphingolipids; Fatty acids and their physicochemical properties; Fats and Waxes - Physicochemical properties and characterization of fats and oils; Structure, Properties and functions of Eicosanoids - Prostaglandins, Prostacyclins, Thromboxanes, Leukotrienes; Chemistry and Properties of Sterols and Steroids – Bile acids and Bile salts; Salient features of Bacterial and Plant lipids

### **Unit – 4**

Nucleic acids: Bases, Nucleosides, Nucleotides; Nucleotides as Energy carriers, Enzyme co-factors and Chemical messengers; Synthetic nucleotide analogs; Chemical synthesis of oligonucleotides; Structure of DNA and different types of DNA, Supercoiled DNA; Structure of RNA and different types of RNA

### **Unit – 5**

Physicochemical properties of Nucleic acids: Denaturation and Renaturation kinetics of nucleic acids - Melting temperature, Cot curves; Sequencing of Nucleic acids – Enzymatic and Chemical methods; Porphyrins – Structure and properties of Porphyrins –Heme, Chlorophylls, Bacteriochlorophylls and Cytochromes

### **Reference books:**

1. Text book of Biochemistry –E.S.West, W.R.Todd et al., 4<sup>th</sup> ed
2. Principles of Biochemistry by Lehninger –D.L.Nelson, M.M.Cox 7<sup>th</sup> ed
3. Text book of Biochemistry with clinical correlations-Thomas M.Devlin, 7<sup>th</sup> ed
4. Harper's review of Biochemistry –D.W. Martin, 19<sup>th</sup> ed
5. Biochemistry – J.M.Berg, J.L.Tymoczko, L.Stryer, 5<sup>th</sup> ed
6. Biochemistry-Reginald H. Garret, Charles M.Grisham 6<sup>th</sup> ed
7. Biochemistry-R.W.McGilvery
8. Biochemistry –J.David Rawn

**M. Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**BC1.2: BIOCHEMICAL TECHNIQUES**

**Course Outcomes:**

- CO1: Enable the students to acquaint with basic principle, instrumentation, procedure, and applications of various classical as well as sophisticated Biochemical techniques
- CO2: Develop competence in various chromatographic techniques and apply them in isolating and characterizing different biological molecules
- CO3: Develop Understanding the principles of Electrophoresis and its applications in biological investigations/experiments and expose the students with various microscopic techniques to study subcellular organelles
- CO4: Provide information on fundamental laws relating to photochemistry and Applications of UV-visible, Fluorescence and IR spectrophotometry in analytical determination and characterization of biomolecules
- CO5: Apply the principles of radiochemistry to analytical determination of biomolecules and life processes

**Course Specific Outcomes:**

- CSO1: Students will be exposed to various chromatographic techniques and their applications in isolation of different biological molecules
- CSO2: In addition to understanding the applications of centrifugation and chromatography in biological investigations, they will gain insight into purification of proteins by affinity chromatography using epitope tags
- CSO3: In addition, Advanced instrumental techniques learned by students may be used for the determination of nutrients, major ions and trace elements, biological samples
- CSO4: The gained knowledge in Photochemistry enables the students to undertake further studies in biochemistry and related areas or in multidisciplinary areas that involve biochemistry
- CSO5: To develop a range of generic skills that are relevant to wage employment, self-employment, and entrepreneurship

**Learning Outcomes:**

After completion of the course, a student will be able to achieve these outcomes.

- LO1: The students will be exposed to different chromatographic techniques like gel filtration, Ion-exchange, thin layer, etc. Students will also learn about various Electrophoretic techniques such as Polyacrylamide Gel Electrophoresis, Agarose Gel electrophoresis etc., and their applications in analysing proteins and nucleic acids
- LO2: The students will learn about Homogenization and Centrifugation techniques
- LO3: The students will also learn the principles of electron microscopy more especially of SEM and TEM and their applications in characterizing biological samples
- LO4: The students will be able to implement the use of instruments like UV-VIS

spectroscopy, NMR, CD, ORD in biological research

LO5: The students will get the theoretical knowledge of various instruments and their practical applications like Geiger-Muller counter, Liquid scintillation counter, Radioactive Isotopes, and safety. X-ray crystallography

### **Unit-1**

Chromatography separation Techniques: Principles, methods, types, and applications of chromatography Techniques – Paper chromatography, Thin layer chromatography, High performance thin layer chromatography, Ion-exchange chromatography, Gel-filtration chromatography and Affinity chromatography: Gas Chromatography, High performance Liquid Chromatography and Chromatofocussing

### **Unit- 2**

Techniques of Electrophoresis: Principles, methods, types and applications of electrophoresis: Moving boundary electrophoresis, Zone electrophoresis, Paper and High voltage electrophoresis Polyacrylamide Gel Electrophoresis, Agarose Gel electrophoresis, Pulsed Field Gel electrophoresis, Isoelectric focusing of proteins, Two-dimensional electrophoresis of proteins, Capillary electrophoresis

### **Unit- 3**

Tissue homogenization Techniques – Disruption of tissues & Cells: Centrifugation Techniques – Basic Principles, instrumentation, types, and applications. Differential and density gradient centrifugation, Preparative and analytical ultra-centrifuge: Principles and applications of Manometer and Oxygen electrode  
Microscopy: Principle, types and applications of microscopes; Phase Contrast, Fluorescent and Electron Microscope

### **Unit- 4**

Spectrophotometry: basic laws of light absorption, instrumentation, and applications of UV –Visible spectrophotometer and IR spectrophotometer  
NMR and ESR spectroscopy, Atomic absorption and Mass spectroscopy,  
Fluorimetry, Flamephotometry and Nephelometry: Optical Rotatory Dispersion (ORD), Circular Dichroism (CD) and X-ray Diffraction

### **Unit-5**

Radioactive isotopes: Nature of radioactivity, Biochemical uses of Radioactive isotopes: Detection and measurements of radioactivity- Liquid scintillating counting, Geiger-Muller counting; Radioactive isotopes and safety- Radiation hazards and methods of radioactive material disposal

### **Reference Books:**

1. Principles and Techniques of Biochemistry and Molecular Biology- K. Wilson, John Walker, 6<sup>th</sup> ed
2. Biophysical chemistry – Upadhyay, Upadhyay, Nath (Himalaya publications)
3. Physical Biochemistry – D. Friefelder, 2<sup>nd</sup> ed
4. Physical Biochemistry – K.E. VanHolde, W.Curtis Johnson et al.,
5. Techniques in Molecular biology- J.M. Walker, Wim Gaastra, vol II
6. An introduction to spectroscopy for Biochemists – S.B. Brown
7. Analytical Biochemistry- David J.Holme, Hazel Peck, 3<sup>rd</sup> ed
8. Lehninger Principles of Biochemistry-David L. Nelson, Michael M. Cox, 7<sup>th</sup> ed
9. Text book of Biochemistry – E.S.West, W.R.Todd et al., 4<sup>th</sup> ed
10. Asokan P, Analytical biochemistry, 2009, Chinna publication

**M.Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**BC 1.3: PHYSIOLOGY AND BIOENERGETICS**

**Course Outcomes:**

- CO1: To understand the role of various physiological processes in the body
- CO2: To know the general mechanism of muscle contraction and nerve impulse transmission
- CO3: To understand the energy transformations in the living system
- CO4: To know the mechanism of ATP synthesis
- CO5: To know the importance of various components involved in photophosphorylation

**Course Specific Outcomes:**

- CSO1: The course highlights the importance of blood clotting and factors involved
- CSO2: The course highlights the role of lungs in exchange of gases and kidneys in acid base balance
- CSO3: Major emphasis was on the mechanism of muscle contraction and nerve impulse transmission
- CSO4: Particular emphasis on understanding the organization of mitochondrial electron transport system and mechanism of ATP synthesis
- CSO5: The course covers the role and importance of photophosphorylation in plants

**Learning Outcomes:**

- LO1: Students will understand various physiological and biochemical processes taking place in the living system
- LO2: Students will acquire a good knowledge in biochemistry of muscle contraction, nerve impulse transmission and biochemistry of vision
- LO3: Students will be aware of free energy transformations, oxidation and reduction reactions that take place in the cells
- LO4: Students will understand the transport of electrons in mitochondria through a series of electron carriers and how ATP synthesis takes place
- LO5: Students will acquire knowledge about absorption of light by chlorophylls and other accessory pigments and mechanism of photophosphorylation

**Unit –1**

Digestion and Absorption of Carbohydrates, Lipids and Proteins

Blood: Composition, properties, and functions - Erythrocytes, Leucocytes, Thrombocytes; Mechanism and Regulation of Coagulation of Blood and Fibrinolysis

Respiration - Mechanism of respiration, Hemoglobin, and transport of gases

Physiology of Heart; Kidney - Structure of kidney and nephron, Regulation of electrolyte, water and acid base balance

**Unit –2**

Muscle Cell: Structure and organization, Types of muscles, Mechanisms of contraction and relaxation of muscle, Biochemical changes associated with muscle contraction and relaxation

Nerve Cell: Structure and organization, Membrane potential, Mechanism of propagation of nerve impulse in unmyelinated and myelinated nerve fibers; Synapse – Types of synapses, Neurotransmitters, Transmission at adrenergic and cholinergic nerve endings; Blood brain barrier; Biochemistry of vision - Rods and Cones, Sensory transduction in vision

### **Unit – 3**

Bioenergetics and Thermodynamics: Principles of thermodynamics - Free energy, Enthalpy and Entropy, Free energy changes in biological transformations in living systems, Equilibrium constant, Coupled reactions; High-energy compounds - Phosphoryl group Transfers and ATP, Thioesters

Biological oxidation and reduction reactions: Electrochemical cell, Nernst Equation, Redox potential; Oxidation and reduction enzymes, Superoxide dismutase, Catalase

### **Unit – 4**

Mitochondrial electron transport system: Organization of components and importance, Q cycle, Thermodynamics of electron transport, Respiratory chain inhibitors, Microsomal electron transport system

Oxidative phosphorylation - Mechanism and theories of Oxidative Phosphorylation, Structure of ATP synthase, Regulation of oxidative phosphorylation – Acceptor control; Uncouplers and Inhibitors of oxidative phosphorylation; Mitochondrial shuttle systems; Substrate level phosphorylation; Bioluminescence

### **Unit – 5**

Photophosphorylation: Light harvesting complexes – Chlorophylls, accessory pigments; Reaction centers -PSI and PS -II -their location; Mechanism of quantum capture and energy transfer between photo systems, Structure of ATP synthase of chloroplasts, Proton gradient and ATP synthesis, Inhibitors of PS-I & PS-II

#### **Reference books:**

1. Textbook of Medical Physiology – A. G. Guyton and J. E. Hall, 10<sup>th</sup> ed
2. Ganong's Review of Medical Physiology - Kim E. Barrett, Susan M. Barman, 29<sup>th</sup> ed
3. Human Physiology - Stuart Fox, 15<sup>th</sup> ed
4. Text Book of Human Physiology – S.Subrahmanyam, K.Madhavankutty
5. Human Physiology – C.C. Chatterjee's, 13<sup>th</sup> ed
6. Human Physiology - Bryan H. Derrickson, 2<sup>nd</sup> ed
7. Biochemistry - Reginald H. Garret, Charles M.Grisham, 6<sup>th</sup> ed
8. Principles of Biochemistry - Lehninger 7<sup>th</sup> ed
9. Principles of Bioenergetics - Skulachev Vladimir P, Alexander V. Bogachev et al.,
10. Bioenergetics: The Molecular Basis of Biological Energy Transformations – Albert L. Lehninger, 2<sup>nd</sup> ed

**M.Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**BC 1.4: ENZYMOLOGY**

**Course Outcomes:**

- CO1: To understand the enzymes, one of the most important types of proteins in the living organisms, enzyme names and commission numbers relate to reactions they catalyze, specific properties
- CO2: To acquire knowledge of biochemical principles with specific emphasis on different metabolic pathways and regulators
- CO3: To gain knowledge on models of enzyme active site, formation of enzyme-substrate complex, enzyme mapping, factors influencing enzyme activity
- CO4: To know about kinetics of enzymatic reactions and different types of enzyme inhibitions explain how enzymes work and behave in living organisms
- CO5: To learn the general principles of mechanism of enzyme catalysis including the role of coenzymes
- CO6: To study the enzyme regulation including allosteric enzymes – characteristics, models, examples and Multi - enzyme systems
- CO7: To understand the molecular basis of various pathological conditions from the perspective of biochemical reactions

**Course Specific Outcomes:**

- CSO1: To understand structure, and functions of enzymes. Learning kinetics of enzyme catalysed reactions and regulatory process, Enzyme activity, Enzyme Units, Specific activity
- CSO2: Have a deeper insight in to the fundamentals of enzyme structure, function and kinetics of enzymes. Discussion on current applications and future potential of enzymes.
- CSO3: Have a complete understanding of rate of reactions and order of reactions. To gain knowledge on enzyme catalysis and isoenzymes and on multienzyme complexes
- CSO4: To learn the models of enzyme action and mapping of enzyme active site
- CSO5: To acquire knowledge on latest concepts of evolution of catalysis
- CSO6: To provide concept of importance of kinetics of enzyme inhibitions and how they influence drug action
- CSO7: To learn the role of enzyme inhibitors in drug discovery and drug design
- CSO8: To gain insight into catalytic mechanisms of enzymes and allosteric regulation of enzymes

**Learning Outcomes:**

- LO1: The students will be able to assign systemic name to enzymes and from the E.C. number they can explain the reaction it catalyzes
- LO2: They will acquire knowledge to analyze the kinetics of different



enzymatic reactions.

LO3: The students will be able to learn how to analyze mechanistic data and they will be able to design experiments to investigate the enzyme inhibition

LO4: They will be able to perform enzyme purification, handling and to characterize new enzymes

LO5: The students will be able to understand the regulation of enzyme activity is so important to coordinate the different metabolic processes and for homeostasis

## **Unit – 1**

Introduction to Enzymes – Nomenclature and Classification of enzymes (IUB), Remarkable properties of enzymes – catalytic power, specificity. Enzyme localization and assay of enzymes, Units of enzyme activity. Active site – Fisher and Koshland models, formation of enzyme – substrate complex and experimental evidences. Nature of active site, mapping of enzyme active site through chemical procedures and site directed mutagenesis. Factors affecting enzyme activity. Modern concepts of evolution of catalysis, ribozymes, abzyme and synzymes

## **Unit – 2**

Enzyme kinetics-Velocity of a reaction, order of a reaction, progress curve for enzyme catalyzed reactions. Kinetics of single substrate enzyme catalyzed reactions, Michaelis – Menten equation, Lineweaver - Burk, Eadie – Hofstee and Hanes plots. Significance of  $V_{max}$ ,  $K_m$ ,  $K_{cat}$ , specificity constant ( $K_{cat}/K_m$ ) Kinetics of multisubstrate reaction – Classification with examples. Rate expression for non-sequential (ping-pong) and sequential (ordered and random) mechanisms. Use of initial velocity, Inhibition and exchange studies to differentiate between multi substrate reaction mechanisms. Flexibility and conformational mobility of enzymes. Reversible inhibition – competitive, non-competitive, uncompetitive and mixed inhibition; Irreversible inhibition -suicide inhibition, Determination of  $K_i$  values

## **Unit – 3**

Strategies of enzyme catalysis – General acid – base, electrostatic, covalent, intermolecular, metal – ion catalysis; Proximity and orientation. Mechanism of reaction catalyzed by serine proteases – trypsin and chymotrypsin; carboxypeptidase, Subtilisin, lysozyme, triose phosphate isomerase, ribonuclease. Rotational catalysis – ATPase

## **Unit – 4**

Role of coenzymes in enzyme catalyzed reactions – Coenzymes and Cofactors. Pyridoxal phosphate, Nicotinamide nucleotides, Flavin nucleotides, Thiamine pyrophosphate, Biotin, Tetrahydrofolate, Pantothenic acid, Cobalamin, Lipoic acid; Metalloenzymes, Isoenzymes-LDH, Creatine phosphokinase. Multifunctional enzymes, properties, Mechanism of action of Pyruvate dehydrogenase and Fatty acid synthase complex

## **Unit-5**

Regulation of enzyme activity – covalent modification, zymogen activity and protein-protein interaction. Allosteric enzymes (ATCase). Cooperativity phenomenon. Hill and Scatchard plots. Sigmoidal kinetics and their physiological significance, Symmetric and sequential models of action of allosteric enzymes and their significance. Feedback inhibition and feed forward stimulation. Control of enzymatic activity by products and substrates. Reversible and irreversible activation. Regulation of Multi-enzyme systems, Pyruvate dehydrogenase and Fatty acid synthase complex

### **Reference books:**

1. Biochemistry - Reginald H. Garret/Charles M. Grisham 6<sup>th</sup> ed
2. Principles of Biochemistry - Lehninger 7<sup>th</sup> ed
3. Understanding enzymes: Palmer T., Ellis Harwood Ltd., 2<sup>nd</sup> ed
4. Enzyme structure and mechanism. Alan Fersht, Freeman & Co 2<sup>nd</sup> ed
5. Enzyme kinetics Siegel interscience – Wiley
6. Lehninger Principles of Biochemistry - David L. Nelson, Michael M. Cox Publisher: W.H. Freeman 8<sup>th</sup> ed
7. Enzyme kinetics - Kent M. Plowman
8. Biological chemistry; H.R. Mehler & E.H Cordes Harper & Rev.
9. Lecture notes on Enzymology - L.G. Whitby, et al.,
10. The Enzyme Molecule - W. Ferdinand
11. Enzymes: Physical Principles - H. Gutfreund
12. Fundamentals of Enzymology, The cell and molecular basis of catalytic proteins - Nicholas C price, Lewis Stevens, 3<sup>rd</sup> ed
13. Enzyme Kinetics: A Modern Approach - Alejandro G. Marangoni, Wiley online library

**M.Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**PRACTICAL – I**  
**BC 1.5: QUALITATIVE ANALYSIS AND BIOCHEMICAL TECHNIQUES**

**Course Outcomes:**

- CO1: To acquire hands on experience to perform general and confirmatory qualitative tests For identification of Carbohydrates (Monosaccharides, Disaccharides, Polysaccharides), Proteins and amino acids, Lipids, Nitrogen bases – Purines, Pyrimidines/Nucleic acids
- CO2: To get hands on experience on various biochemical techniques such as Paper chromatography, Thin layer chromatography, Ion-exchange chromatography, Affinity chromatography, Gel filtration, Paper electrophoresis, SDS-PAGE, 2D Electrophoresis, Differential and Density gradient centrifugation, and handling Colorimeter, Spectrophotometer, and Polarimeter

**Course Specific Outcomes:**

- CSO1: Students exposure to basic reactions of biomolecules
- CSO2: To gain knowledge to determine presence of biomolecules like carbohydrates, proteins, lipids etc. in known and unknown samples
- CSO3: The students should be able to determine the extent of adulteration in samples containing Biomolecules
- CSO4: The students should obtain hands-on training in basic separation techniques in biochemistry like chromatography, electrophoresis, etc.
- CSO5: They gain expertise in the isolation of various biomolecules and organelles
- CSO6: To develop competence in applying various chromatographic techniques in isolating and characterizing different biological molecules
- CSO7: Understanding the principles and applications of chromatography, centrifugation, electrophoresis, spectrophotometry and ELISA tools in biological investigations/experiments

**Learning Outcomes:**

- LO1: The students will be able to learn to identify carbohydrates, proteins, amino acids, lipids, nitrogen bases - purines, pyrimidines/Nucleic acids from a given unknown sample
- LO2: The students will get the expertise for analysis of any biological or non biological sample for identification of its chemical composition
- LO3: The students will develop skills to perform various Biochemical Techniques such as Paper chromatography, Thin layer chromatography, Ion-exchange chromatography, Affinity chromatography, Gel filtration, Paper electrophoresis, SDS-PAGE, 2D Electrophoresis, Differential and Density gradient centrifugation, and their applications for separation of different biomolecules and isolation of cells and tissues for studying their functional abnormalities

- LO4: The students will be able to learn techniques for isolation, purification and chemical characterization of compounds from plants and microbes which will have medical or commercial importance
- LO5: The students will be able to measure and detect the biomolecules and molecules involved in a reaction using the colorimeter
- LO6: The students will be able to learn application of UV-visible spectroscopy
- LO7: The students will learn different methods of protein estimation
- LO8: The students will be able to learn the principles of Peptide mapping and Isoelectric focusing
- LO9: The expertise gained by the student from this practical experiments can be useful in Pharmaceutical and Biotech industries

### **QUALITATIVE ANALYSIS (A)**

General reactions of Carbohydrates  
 Reactions of Polysaccharides  
 General colour reactions of proteins  
 Identification of unknown sugars  
 Identification of unknown amino acids  
 General reactions of lipids  
 Reactions of cholesterol  
 Identification of unknown lipids  
 General reactions of nitrogen bases  
 Reactions of nucleic acids  
 Identification of unknown nitrogen bases/ nucleic acids

### **BIOCHEMICAL TECHNIQUES (B)**

Separation of amino acids by ascending paper chromatography  
 Separation of amino acids by descending paper chromatography  
 Separation of sugars by ascending paper chromatography  
 Separation of nucleic acids by ascending paper chromatography  
 Separation of amino acids by thin layer chromatography  
 Separation of lipids by thin layer chromatography  
 Separation of plant pigments by column chromatography  
 Separation of amino acids by ion exchange chromatography  
 Affinity Chromatography  
 Separation of amino acids by paper electrophoresis  
 Separation of serum proteins by Polyacrylamide Gel Electrophoresis  
 Molecular weight of protein by SDS-PAGE  
 Absorption spectrum of chlorophyll extracted from green leaves  
 Absorption spectrum of aromatic amino acids, purines, pyrimidines and heme  
 Estimation of proteins by Spectrophotometric method  
 Determination of Molar absorption coefficient of tyrosine  
 Sub – cellular fraction of organelles of liver cells and identification by the marker enzymes.  
 Optical rotation of glucose and fructose using polarimeter.

N and C terminal analysis of proteins. (End group analysis of proteins)

Peptide mapping

Density gradient centrifugation – Isolation of rat liver mitochondria

2- Dimensional electrophoresis of proteins

Isoelectric focussing

**Reference books:**

1. Experimental Biochemistry-B.Sashidhar Rao, Vijay Deshpande
2. Principles and Techniques of Practical Biochemistry-Wilson and Walker
3. An introduction to Practical Biochemistry-David T.Plummer, 3<sup>rd</sup> ed
4. Laboratory Manual in Biochemistry-J.Jayaraman
5. Principles and Techniques of practical Biochemistry. Eds. Williams and Wilson.
6. A Biologists guide to Principles and Techniques of practical Biochemistry  
– Ed.Bryan, L.Williams& Keith Wilson (Edward Arnold).

**M.Sc. BIOCHEMISTRY**  
**I SEMESTER**  
**PRACTICAL – II**  
**BC 1.6: ENZYMOLOGY I AND ENZYMOLOGY - II**

**Course Outcomes:**

- CO1: To become familiar with the basic methods of studying enzymes
- CO2: To understand the mechanism of action of enzymes
- CO3: To get familiar with the kinetics of enzyme reactions
- CO4: To apply appropriate methods for determination of catalytic parameters
- CO5: To adopt the systematic procedure to purify enzymes

**Course Specific Outcomes:**

- CSO1: The course gives in depth knowledge in understanding the mechanism of enzyme action
- CSO2: The effect of parameters like  $P^H$ , temperature on enzyme activity were thoroughly understand by students
- CSO3: To understand the effect of inhibitors on the mechanism of enzyme action

**Learning Outcomes:**

- LO1: Students will acquire practical knowledge on isolation of enzymes from different sources, handling and preservation of enzymes without losing their activity
- LO2: Students will learn how to calculate enzyme activity and how the velocity of the reaction changes in presence of inhibitors
- LO3: Students will learn different techniques for purification of enzymes which enable them to use these techniques in future research and also to find appropriate jobs in scientific-research laboratories
- LO4: Students will gain knowledge about applying enzymes and their inhibitors in medicine and various industries

**ENZYMOLOGY - I (A)**

Assay of Amylase from saliva

Assay of Acid phosphatase from potato

Assay of Trypsin

Assay of urease from Horse – gram

Assay of Succinate dehydrogenase from the liver

Enzyme purification by 3 or 4 steps

- a) Acetone precipitation
- b) Ammonium sulphate fractionation
- c) Ion – exchange chromatography
- d) Gel filtration
- e) Electrophoresis

Isoenzymes of LDH – electrophoretic separation and specific staining technique

## ENZYMOLGY - II (B)

Time course of enzyme activity

Effect of pH on enzyme activity and determination of optimum pH

Effect of temperature on enzyme activity and calculation of energy of activation Effect of substrate concentration on enzyme activity and determination of Michealis constant

Effect of metal ions on enzyme – Alcohol dehydrogenase

Effect of substrate and regulators on allosteric enzyme – Phosphorylase or ATCase

Enzyme inhibition – irreversible inhibition of Papain or Serine proteases by appropriate inhibitors

### Reference books:

1. An introduction to Practical Biochemistry-David T.Plummer, 3<sup>rd</sup> ed
2. Hans Bisswanger, Practical Enzymology, 2nd Edition, Wiley Online Library
3. Enzymes: A Practical Introduction to Structure, Mechanism, and Data Analysis- - Robert A. Copeland, 2<sup>nd</sup> ed