# M.Sc.-Biotechnology Semester –I BIT070: Biochemistry and Metabolism [Credit: 6(4+2)]

# UNIT I

**Bioenergetics:** Stabilizing interactions (Van der Waals, electrostatic, hydrogen bonding, hydrophobic interaction, etc.) Laws of thermodynamics, Gibbs free energy, endergonic & exergonic reactions, Standard state free energy changes, High energy compounds. Introduction to Metabolism - Catabolism, anabolism, catabolic, anabolic and amphibolic pathways.

# UNIT II

**Carbohydrates:**Structure and properties of mono, oligo and polysaccharides. Metabolism and regulation-Glycolysis, TCA cycle, Glyoxylate cycle. Pentose phosphate pathway, Gluconeogenesis, Entner – Doudoroff pathway. Substrate level phosphorylation; Oxidation- Reduction reactions. Redox potential, Electron transport chain, Oxidative phosphorylation and ATP synthesis.

# UNIT III

**Lipids:** Classification, structure of saturated and unsaturated fatty acids, triacylglycerol, phospholipids, glycolipids and sterols; Oxidation of fatty acids ( $\alpha$ ,  $\beta$ ,  $\omega$  oxidation). Biosynthesis of fatty acids (saturated and unsaturated) and sterol. Ketone bodies synthesis.

## UNIT IV

Amino acids & proteins: Classification, structure and properties of amino acids. General aspects of amino acid metabolism; amination, transamination, deamination. Decarboxylation, urea cycle. Classification, properties and structural organization of proteins- primary structure, secondary structure, Ramachandran plot, tertiary structure and quaternary structure.

## UNIT V

**Nucleic acids:** Structure of bases, nucleosides and nucleotides; Conformation of nucleic acids: helix (A, B, Z), t-RNA, micro-RNA. Biosynthesis: Purine and pyrimidine, *denovo* and salvage pathway.

## PRACTICALS

- 1. Concept of pH and buffers, preparation of buffers.
- 2. Estimation of protein by Bradford method
- 3. Estimation of protein by Lowry method
- 4. Estimation of reducing sugars by DNS Method
- 5. Estimation of DNA by DPA Method
- 6. Estimation of RNA by Resorcinol Method
- 7. Isolation of lipolytic microbes from soil plate method and estimation of total lipids
- 8. Fractionation of total lipids (glycolipids, neutral lipids and phospholipids) by column chromatography.
- 9. Extraction and estimation (by TLC) of ergosterol.
- 10. Sugar estimation by thin layer chromatography.
- 11. Lipid estimation by thin layer chromatography.
- 12. Separation of amino acids by paper chromatography.

- 1. Voet, D. and Voet, J.G. (2004). Biochemistry, John Wiley and Sons.
- 2. Roger, L.P., Adams, Johm T., Knowler and David P., Leader. (1992). The Biochemistry of the Nucleic Acids. 11th edition. Chapman and Hall.
- 3. Smith and Wood (1991). Energy in Biological Systems. Chapman and Hall.
- 4. Berg, J. M., Tymoczko, J. L. and Stryer, L. (2006). Biochemistry. VI Edition. W.H Freeman & Co.
- 5. Buchanan, B., Gruissem, W. and Jones, R. (2000) Biochemistry and Molecular Biology of Plants. American Society of Plant Biologists.
- 6. Nelson, D.L., Cox, M.M. (2004) Lehninger Principles of Biochemistry, 4th Edition, WH Freeman and Company, New York, USA.
- 7. Hopkins, W.G. and Huner, P.A. (2008) Introduction to Plant Physiology. John Wiley and Sons.
- 8. Salisbury, F.B. and Ross, C.W. (1991) Plant Physiology, Wadsworth Publishing Co. Ltd.

# M.Sc.-Biotechnology Semester –I BIT080: Functional Cell Biology [Credit: 6(4 +2)]

# **UNIT I: Cellular Organization**

Structure, organization and composition of prokaryotic and eukaryotic cell. Plasma membrane structure and functions, membrane models. Structural organization and function of intracellular organelles- Cell wall, nucleus, mitochondria, Golgi bodies, lysosomes, endoplasmic reticulum, peroxisomes, plastids, vacuoles, chloroplast.

### **UNIT II: Cytoskeleton**

Nature of cytoskeleton, Actin filaments, Actin binding proteins, Intermediate filaments, Microtubules, MAPs, Structure and functions of cilia and flagella.

### **UNIT III: Membrane Transport**

Transport across membrane- passive diffusion, osmosis, active transport, Ion Channels, ABC transporters,  $Na^+$  and  $K^+$  pump,  $Ca^{2+}$  ATPase pump, co-transport, symport, antiport, endocytosis and exocytosis. Membrane vesicular traffic.

## **UNIT IV: Cell Signaling**

Cell to cell interactions, Cell adhesion-integrins, selectins, cadherins. Cell Junction- Tight and gap junctions, Desmosomes, plasmodesmata. extracellular matrix. General principles of cell signaling, cell surface receptor, signaling via G-protein coupled receptors, signal transduction pathways, role of secondary messengers, regulation of signaling pathways.

#### **UNIT V: Cell Cycle and Cancer**

Molecular events of cell division and cell cycle, regulation of cell cycle events- Cyclins, Cyclin dependent kinases, inhibitors. Introduction to cancer, oncogenes, tumor suppressor genes, cancer and the cell cycle, virus-induced cancer, metastasis, interaction of cancer cells with normal cells, apoptosis, necrosis.

### PRACTICAL'S

- 1. Study of mitosis and meiosis.
- 2. Isolation of mitochondria
- 3. Isolation of Chloroplast
- 4. Isolation of genomic DNA from different sources.
- 5. RNA isolation from yeast cells
- 6. Quality and quantity checking of DNA by UV spectrophotometer
- 7. Quality and quantity checking of RNA by UV spectrophotometer
- 8. Microtomy: Fixation, block making, section cutting and staining of tissues
- 9. Study of the Barr body from the (female) smear of Buccal epithelial cells.

- 1. Freifelder. D. (2003) Essentials of molecular Biology fourth edition, Jones and Bartlett Publications Inc.
- 2. Lewin.B. (2007) Genes IX, Jones and Bartlett Publishers
- Turner, P.C., Mclennan, A.D. Bates, A.D. (2005) Instant notes Molecular Biology III Edition, Routledge, UK 6
- Watson, J.D. (1987) Molecular Biology of Gene The Benjamin / Cummings Publishing Company Inc., California
- 5. Darnell, J.E., Lodish, H, and Baltimore, D. (2000) Molecular Cell Biology, Fourth Edition, W.H. Freeman and Company, New York.
- 6. Stanley. R. Maloy. John.E. Cronan., David Freifelder (1998), Microbial Genetics, II edition, Narosa Publishing House, Madras
- 7. Strickberger (1996), Genetics, Prentice Hall of India Pvt. Ltd., New Delhi
- 8. Brown. T.A. (2006), Genomes 3, Garland Science Publications
- 9. Bruce Alberts, Alexander Johnson, Julian Lewis, Martin Raff. Keith Roberts, Peter Walter, (2002), Molecular Biology of the Cell, IV edition, Garland Publishing, New York
- 10. Lodish, Harvey, Arnold, Matsudaira, Paul, Kaiser, Chris. A, Krieger, Monty Scott, Matther P. Zipuruky, Lawrence, Darnell, James (2004), Molecular Cell Biology, W.H. Freeman & Company

# M.Sc.-Biotechnology Semester –I BIT090: Microbiology [Credit: 5(3+2)]

### **UNIT I: Microbial Systematic classification**

Classical and modern methods and concepts; Binomial nomenclature, Whittaker's five kingdom and Carl Woese's three kingdom classification systems and their utility. Introduction of Bergey's manual of bacteriological classification. 16s rRNA sequence and bacterial phylogeny.

## **UNIT I: Microbial Diversity**

Bacteria: General characteristics with emphasis on their morphology and cell structure. Algae: General structure and characteristics, Applications in biotechnology. Fungi and slime moulds: General structure and characteristics, applications in biotechnology. Viruses: General structure and characteristics of acellular microorganisms (Viruses, Viroids, Prions), Life cycles: T4 and lamda phage.

## UNIT III: Microbial growth and nutrition

Definitions of growth, measurement of microbial growth, Batch culture, Continuous culture, generation time and specific growth rate, synchronous growth, diauxic growth curve. Microbial growth in response to environment: Temperature (psychrophiles, mesophiles, thermophiles, extremophiles, thermodurics, psychrotrophs), pH (acidophiles, alkaliphiles), solute and water activity (halophiles, xerophiles, osmophilic), Oxygen (aerobic, anaerobic, microaerophilic, facultative aerobe, facultative anaerobe), barophilic. Microbial growth in response to nutrition and energy: Autotroph/Phototroph, heterotrophy, Chemolithoautotroph, Chemolithoheterotroph, Chemolithotroph, photolithoautotroph and Photoorganoheterotroph.

## **UNIT IV: Microbial Control**

Fundamentals of control: the rate of death of bacteria, conditions influencing antimicrobial action, mode of action of antimicrobial agents. Physical methods of sterilization: Moist Heat, Dry Heat, Tyndallization, Filteration, radiation, osmotic pressure, desiccation. Chemical methods of microbial control: sanitizers, disinfectants, antiseptics, antimicrobial action of phenols and phenolic compounds, alcohols, halogens, heavy metals, dyes, aldehydes, detergents. Antibiotics and their mode of action.

#### **UNIT IV: Microbial genetics**

Types of plasmids – F plasmid, R Plasmids, colicinogenic plasmids. Transformation- Discovery, mechanism of natural competence. Conjugation- Discovery, mechanism, Hfr and F' strains, Interrupted mating technique and time of entry mapping. Transduction- Generalized transduction, specialized transduction, LFT & HFT lysates.

# PRACTICALS

- 1. Isolation of microorganism by plating, streaking & serial isolation methods.
- 2. Maintenance of cultures by slants & stab method, storage of microorganism
- 3. Microscopic observation Gram staining, Capsule & Spore Staining
- 4. Study of Diauxic growth curve
- 5. Effect of environmental factors on growth of bacteria: Salt, Temp, pH.
- 6. Viable count of bacteria from soil sample (Dilution Plating Method)
- 7. Biochemical characterization of selected Microbes
- 8. Isolation of bacteriophages from sewage sample
- 9. Enrichment and Isolation of: a) Halophiles b) Acidophiles c) Phenol Degraders d) Nitrogen Fixers e) Antibiotic Producers f) Kojic Acid Producers
- 10. Effect of Antibiotics on various bacteria
- 11. Determination of minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of various antibiotics on different organisms

- 1. Pelczar M.J.Chan, 5th Edition, Microbiology
- 2. Prescott L. M. Microbiology, 6th Edition
- 3. Jhonson ,Laboratory Experiments in Microbiology,6th Edition, Pearson Education
- 4. Singleton Sainsbury, Dictionary of Microbiology & Molecular Biology, John Wiley
- 5. R.C. Dubey& Maheshwari, A Textbook of Microbiology, 1st Edn, 2005.
- 6. Medical Microbiology, Anantnarayan
- 7. Torotora, Microbiology: An Introduction 8th Edition.

# M.Sc.-Biotechnology Semester –I BIT100: Enzymology and Bioinstrumentation [Credit: 5(3+2)]

### **UNIT I: Enzyme – General Account**

Definition, Classification of enzymes, specificity, active sites, coenzymes, enzyme units, isozymes, Mechanism of enzyme action- lock and key, and induced fit hypothesis, acid-base, covalent and metal ion catalysis. Factors affecting rate of enzymatic reactions: temperature, pH, modulators etc. and significance of activation energy and free energy in biochemical reactions. Ribozymes and abzyme.

### **UNIT II: Enzyme Kinetics**

Michaelis-Menten hypothesis, Transformation of Michaelis- Menten equation and determination of Km and Vmax, Significance of Km and Vmax, Lineweaver–Burk plot, Determination of kinetic parameters, multi substrate kinetics. Haldane relationship, Hills equations. Enzymes inhibition: Covalent, allosteric and feedback inhibition; Reversible (competitive, noncompetitive and uncompetitive) and irreversible inhibitions.

## **UNIT III: Protein Engineering**

Concept and Methods, Site directed mutagenesis, Active site mapping, Nature of the active site, Identification of functional groups at the active site, Enzyme immobilization - types, advantages, drawbacks and applications; Artificial enzymes; Isolation and purification of industrially important enzymes.

# UNIT IV: Enzyme technology for industrial application

Large scale production of enzymes, Applications of enzyme technology in environment, Medical, Agricultural, food and chemical industries.

## **UNIT V: Bioinstrumentation**

Microscopy: Simple microscopy, phase contrast microscopy, florescence and electron microscopy (TEM and SEM), absorption and emission spectroscopy. Chromatography: paper chromatography, thin layer chromatography, gel filtration, affinity and ion exchange Chromatography, gas chromatography, HPLC, HPTLC. Spectrometry: UV-Vis, atomic absorption spectrophotometer. Electrophoresis: Agarose gel, native and SDS PAGE.

# PRACTICALS

- 1. Isolation of extra cellular enzymes.
- 2. Isolation of intra cellular enzymes.
- 3. Enzyme kinetic (Vmax and Km values) study of amylase
- 4. Enzyme assay: protease, lipase
- 5. Enzyme purification: ammonium sulphate method
- 6. Agarose gel electrophoresis
- 7. Molecular weight determination of enzyme by electrophoresis
- 8. Quantitative estimation of enzyme using UV-Vis spectrophotometer
- 9. To perform enzyme immobilization
- 10. Separation of analytes using gel filtration chromatography
- 11. Separation of analytes using ion exchange chromatography

- 1. Biological chemistry, H.R Mahier & E. Cordes 1986.
- 2. Enzymes, Dizon & Webb.
- 3. Genes VII, Benjemin Lewin, 1994. Oxford University Press. Oxford
- 4. Principles of Biochemistry, AL. Lehninger, D.L. Nelson and M. M. Cox. 1993. Worth Publishers, New York.
- 5. Palmer, T. (2001). Enzymes: Biochemistry, Biotechnology and Clinical Chemistry, Horwood Publishing Chichester.