

**DEPARTMENT OF BIOCHEMISTRY & BIOTECHNOLOGY**  
**M.Sc. BIOTECHNOLOGY**  
**Choice Based Credit System (CBCS)– 2 year program (2018-2019)**  
**Revised Scheme of Examinations and Syllabus**

Subject Code	Theory & Practical	Credit			Internal Assessment Marks	End Semester Examination Marks	Total Marks
		L	P	C			
<b>SEMESTER-I</b>							
BIT C 101	Basic Biochemistry	4	-	4	25	75	100
BIT C102	Cell Biology and Genetics	4	-	4	25	75	100
BITP 103	Practical I	-	12	4	40	60	100
	Optional-I	4	-	4	25	75	100
ENG C 116	Soft Skills	4	-	4	25	75	100
<b>Total Credits: 20</b>							
<b>SEMESTER-II</b>							
BIT C 201	Enzyme Technology	4	-	4	25	75	100
BIT C 202	Immunology & Immunotechnology	4	-	4	25	75	100
BIT C 203	Industrial and Environmental Biotechnology	4	-	4	25	75	100
BIT C 204	Molecular Biology	4	-	4	25	75	100
BIT P 205	Practical II	-	9	3	40	60	100
BIT P206	Practical III	-	9	3	40	60	100
	Optional-II	4	-	4	25	75	100
<b>Total Credits: 26</b>							
<b>SEMESTER –III</b>							
BIT C 301	Analytical Techniques	4	-	4	25	75	100
BITC 302	Genetic Engineering and Nanobiotechnology	4	-	4	25	75	100
BIT C 303	Plant Biotechnology	4	-	4	25	75	100
BIT C 304	Animal Biotechnology	4	-	4	25	75	100
BIT P 305	Practical IV	-	12	4	40	60	100
	Optional-III (Value Added Course)	4	-	4	25	75	100
<b>Total Credits: 24</b>							
<b>SEMESTER IV</b>							
BIT C 401	Genomics , Proteomics and Bioinformatics	4	-	4	25	75	100
BIT C 402	Food and Medical Biotechnology	4	-	4	25	75	100
BIT P 403	Practical V	-	10	3	40	60	100
	Optional –IV (Value Added Course)	4	-	4	25	75	100
BIT PJ 404	Project	-	-	5	25	75	100
<b>Total Credits:20</b>							

**Distribution of Credits**

Core Courses and Project : **70 credits**  
Soft Skills and Optional : **20 credits**  
**Total : 90 credits**

Optionals: Any 4 from the following:

1. Chemistry
2. Microbiology
3. Statistical Methods
4. Drug Design and Drug Action
5. Genetic Engineering
6. Yoga
7. Vermiculture & Sericulture
8. Marine Biotechnology
9. Biotechnology Management
10. Clinical Biochemistry
11. Any other course offered by other departments from time to time.

AIDS. Antisense and siRNA therapy. Nanotherapy. Stem cell therapy. Bioethics- Food and drug safety. Ethical issues in human gene therapy, human genome analysis and human cloning.

#### **Text Books**

1. Mathews & Montville et al. Food Microbiology: An introduction. 4<sup>th</sup> ed. ASM Press. 2017.
2. Borem et al Understanding Biotechnology. Pearson 2011.
3. Adams and Moss. Food Microbiology. 4<sup>th</sup> ed. Royal SocChem 2015.
4. Jay et al. Modern Food Microbiology 7<sup>th</sup> ed. Springer 2006.
5. Glick and Pasternak. Molecular Biotechnology. 4<sup>th</sup> ed. ASM Press 2009.
6. Singh BD. Biotechnology. Kalyani Publ.2004

#### **Reference Books**

1. Ward OP. Fermentation Biotechnology. John Wiley 1991.
2. Maulik and Patel Molecular Biotechnology Wiley-Liss. 1997.
3. Watson et al. Recombinant DNA: Genes and genomes - A short course. 3<sup>rd</sup> ed. Freeman 2006.

#### **BITP 403: PRACTICAL V Bioinformatics, Food and Medical Biotechnology**

1. Sequence alignment and searching
2. Gene prediction
3. Multiple sequence alignment
4. Phylogenetic analysis
5. Protein sequence analysis, structure prediction
6. Primer design
7. SNP and ORF finding in DNA sequence
8. Visualization tools.
9. Molecular modeling.
10. Dehydration of fruits and vegetables.Preparation of fruit juice powders.
11. Isolation of microbes from spoiled vegetables.
12. Preparation of fruit juice concentrates and use of enzymes for clarity.
13. Identification & characterization of proteins resolved on 2D PAGE Demo.
14. HPTLC and GC-MS- Demonstration.
15. Structure determination of proteins and nucleic acids by NMR & XRD-Demo.
16. Aseptic packaging, freeze preservation, drying and dehydration, food fermentation, pickling and curing.
17. Preservation of food products using chemical preservatives.
18. Tissue collection, formalin fixation, sectioning, and staining.
19. Analysis of biochemical analytes by autoanalyser.
20. Biochemical analyses for diabetes (blood glucose) and cardiovascular disease (serum cholesterol).
21. Use of ELISA for disease diagnosis- demonstration.

## BITC 402: FOOD AND MEDICAL BIOTECHNOLOGY

**Objective:** To acquire knowledge in food biotechnology, molecular basis of diseases, molecular diagnostics & therapeutics.

### Unit-I Food spoilage and preservation

Types and sources of microorganisms associated with food. Conditions influencing microbial growth in food. Composition and spoilage of food, meat, fish, cereals, pulses, nuts and oil seeds, fruits and fruit products, vegetable and vegetable products. Methods of food preservation. Control of microorganisms by retarding growth- low temperature, drying, intermediate moisture, chemicals. Control of microorganisms by destruction- gas treatments, heat, ionization radiation, ultraviolet radiation. canning and packing (Elementary idea).

### Unit-II Fermented foods and enzymes in food industry

Basic principles of food fermentation. Fermented foods: fermented milk- yoghurt, cheese, bread; fermented vegetables- sauerkraut, olives. Fermented meats and fish. Production of beer, wine, and vinegar. Mushroom farming. Use of enzymes in food industry- proteases in food processing, enzymes in baking and dairy industry, enzymes in fruit juice and brewing industry. Pickling and curing.

### Unit-III Molecular Basis of diabetes, atherosclerosis & cancer

Role of tissues and hormones in blood sugar homeostasis. Diabetes mellitus: classification, diagnosis, management, complications. Atherosclerosis: risk factors and management. Cancer- differences between benign and malignant tumours, growth characteristics of cancer cells, mechanism of radiation, virus and chemical carcinogenesis. Oncogenes and tumor suppressor genes (brief account).

### Unit-IV Molecular Diagnostics

Diagnostic kits. Tumor markers- oncofetal proteins, hormones, enzymes, tumor-associated antigens. Prenatal & neonatal screening for genetic disorders. DNA diagnostic systems- probes. RFLP & PCR in disease diagnosis. Histocompatibility testing: cross matching. Viral diagnostics: immunodiagnosis, molecular diagnosis. SNP-based diagnosis.

### Unit-V Molecular Therapeutics

Mabs, growth factors and interferons as therapeutic agents. Therapeutic agents from nonrecombinant and recombinant organisms. Antivirals and antiretrovirals. Drug delivery and targeting. Gene therapy: gene delivery systems, *ex vivo* and *in vivo* strategies, gene therapy for single-gene disorders, cancer and

## BITC 101: BASIC BIOCHEMISTRY

**Objective:** To comprehend the structure-function relationships of various biomolecules and concepts of metabolism.

### Unit-I Bioenergetics and Biological Oxidation

Laws of thermodynamics. Basic concepts of free energy, entropy and enthalpy. Standard free energy change. Exergonic and endergonic reactions. Bioenergetics: high energy phosphate compounds, the ATP/ADP cycle. Synthesis, utilization and breakdown of ATP. Biological oxidation: Redox reactions and oxidoreductases. Electron transport chain: components, role in energy capture, respiratory control. Oxidative phosphorylation- Chemiosmotic theory, inhibitors, uncouplers and ionophores. Introduction to metabolism - anabolism and catabolism.

### Unit-II Carbohydrates and Lipids

Classification of carbohydrates. Biologically important monosaccharides and disaccharides. Structure and biological functions of homopolysaccharides and heteropolysaccharides. Carbohydrate metabolism (structures not required)- brief outline of glycolysis, citric acid cycle, gluconeogenesis, pentose phosphate pathway, glycogen metabolism.

Classification of lipids. Fatty acids. Biological functions of eicosanoids, phospholipids, sphingolipids, and cholesterol. Lipid metabolism (structures not required): brief outline of fatty acid oxidation and lipogenesis.

### Unit-III Proteins-I

Amino acids: 3-letter and 1-letter abbreviation for amino acids. Classification and general properties. Biologically important peptides. Proteins: classification, denaturation and renaturation. Orders of protein structure: Primary structure- determination of the amino acid sequence of proteins. The peptide bond, Ramachandran plot. Secondary structure:  $\alpha$ -helix,  $\beta$ -sheet and  $\beta$ -turns. Pauling and Corey model.

### Unit-IV Proteins-II

Supersecondary structure- helix-loop-helix, hairpin  $\alpha$  motif,  $\beta$ - $\alpha$ - $\beta$  motif. Tertiary structure-  $\alpha$  and  $\beta$ -domains. Quaternary structure of proteins. The structure of hemoglobin.

Protein metabolism (structures not required): Catabolism of amino acid nitrogen- transamination, deamination, ammonia

formation, urea cycle, catabolism of carbon skeletons (overview only). Conversion of amino acids to specialized products.

### **Unit–V Nucleic acids and Nucleic acid-binding proteins**

Nitrogenous bases, nucleosides and nucleotides. Biologically important nucleotides.

DNA double helical structure- Watson and Crick model. A, B, and Z forms of DNA. Triple and quadruple structures. DNA supercoiling and linking number. Properties of DNA: buoyant density, viscosity, denaturation, cot curve. Differences between DNA and RNA. Major classes of RNA- structure and biological functions. Minor classes of RNA.

DNA binding proteins- the helix-turn-helix motif, zinc finger, leucine zipper, and helix-loop-helix.

#### **Text Books**

1. Nelson and Cox. Lehninger Principles of Biochemistry. Freeman, 7th ed. 2017.
2. Voet and Voet. Fundamentals of Biochemistry. 5th ed. Wiley. 2016.
3. Murray et al. Harper's Illustrated Biochemistry 30th ed. McGraw Hill, 2015.
4. Berg, Tymoczko. Stryer Biochemistry 8th ed. Freeman 2015.

#### **Reference Books**

Blackburn et al. Nucleic acids in Chemistry and Biology. Royal Soc Chem 2006.

## **BITC 102: CELL BIOLOGY AND GENETICS**

**Objective:** To learn in detail about the molecular organization of cells and cellular processes and the principles of genetics.

### **Unit–I Cell and tissue organization**

Prokaryotic and eukaryotic cells. Structure and functions of subcellular organelles. The cytoskeleton- microtubules, microfilaments and intermediate filaments. Types of tissues. Epithelium- organization and types. The basement membrane. Connective tissue.

Major classes of cell junctions- anchoring, tight and gap junctions. Major families of cell adhesion molecules (CAMs)- cadherins, integrins. Brief account of the extracellular matrix.

## **Unit–IV Structural & functional proteomics & applications**

Structural proteomics: X-ray and NMR for protein structure analysis. Comparative and homology modeling, secondary structure prediction, fold recognition and *ab initio* prediction. SCOP. Protein sequence analysis: substitution score matrices, pairwise similarity search, pattern recognition. Protein function determination: database search for homology, phylogenetic profile method, domain fusion. Protein-protein interactions: yeast 2-hybrid system. Protein arrays and chips (concept and applications). Applications of proteomics.

### **Unit–V Bioinformatics**

Useful search engines. File formats. PubMed. Bioinformatics workstation, Unix. Biological databases (primary, secondary, organism-specific, miscellaneous). Data submission and retrieval. Sequence alignment: substitution scores and gap penalties. Database similarity searching: BLAST, FASTA. Multiple sequence alignments: CLUSTAL. Gene discovery and prediction. Molecular phylogenetics: phylogenetic tree construction and analysis. Identification of orthologs and paralogs. Protein structure database- protein structure visualization, comparison and classification. Protein motifs and domain prediction.

#### **Text Books**

1. Lesk A. Introduction to Genomics. 4<sup>th</sup> ed. Oxford Univ Press. 2013.
2. Primrose. Principles of genome analysis. 3<sup>rd</sup> ed. Wiley 2002.
3. T. A. Brown. Genomes. 2007, 4<sup>th</sup> ed Garland Science.
4. Dale and von Schantz. From Genes to Genomes: Concepts and applications of DNA technology. 3rd ed. Wiley-Interscience. 2011.
5. Lovrik Introducing Proteomics. Wiley-Blackwell. 2011.
6. Twyman. Principles of Proteomics. 2<sup>nd</sup> ed. 2013
7. Liebler DC. Introduction to proteomics. Humana Press. 2<sup>nd</sup> ed. 2009.
8. Hodgman et al. Instant Notes in Bioinformatics. 2<sup>nd</sup> ed. Taylor and Francis, 2009.

#### **Reference Books**

1. Gibas and Per Jambeck. Developing bioinformatics computer skills. 2<sup>nd</sup> ed. O'Reilly Associates, 2013.
2. Baxevanis, Ouellette. Bioinformatics. A practical guide to the analysis of genes and proteins. 3<sup>rd</sup> ed. Wiley Interscience, 2004.

4. Separation of plant pigments by adsorption chromatography.
5. Separation of proteins by SDS-PAGE.
6. HPLC-Demonstration.
7. Subcellular fractionation.
8. Plasmid preparation, characterization by electrophoresis.
9. Restriction enzyme digestion of DNA.
10. PCR and analysis of PCR products
11. Blotting techniques - Western.
12. GFP cloning. Demonstration
13. Protoplast isolation and culture.
14. Preparation of tissue culture medium and membrane filtration.
15. Preparation of single cell suspension, cell counting, viability of cells.
16. Trypsinization of monolayer and subculturing.
17. MTT assay for cytotoxicity testing.

### **BITC 401: GENOMICS, PROTEOMICS, AND BIOINFORMATICS**

**Objective:** To understand the principles of genome mapping, sequencing, and genome analysis, and the tools and applications of proteomics and bioinformatics.

#### **Unit-I Genome mapping and sequencing**

Genome mapping-rationale. Types of gene map-genetic, cytogenetic and physical. Molecular markers for mapping-RFLPs, microsatellites and SNPs. Assembling a physical map of the genome - chromosome walking and jumping. Genome sequencing approaches: whole-genome shotgun, hierarchical shotgun. Identifying genes- sequence inspection, EST comparison, similarity searches.

#### **Unit-II Genome projects, post-genome analysis**

Genome projects: genome sequence data of model organisms- *E.coli*, *D.melanogaster*, and mouse. The Human Genome Project: goals, mapping strategies, markers, sequencing technologies, results of final sequence, potential benefits and risks, ELSI. Post-genome analysis- differential display, DNA microarray, ChIPs, knock-out analysis.

#### **Unit-III Protein separation, identification and quantitation**

Proteomics-introduction. Protein separation- general principles. 2D-gel electrophoresis, liquid-liquid chromatography. Protein identification by antibodies, Edman degradation, mass spectrometry-basic principle and instrumentation, ESI, MALDI-TOF, SELDI-TOF, tandem MS. Peptide mass fingerprinting (elementary details).

### **Unit – II Membrane composition and transport**

Composition of membranes- the lipid bilayer, peripheral and integral proteins. The fluid mosaic model. Brief account of membrane rafts. Endocytosis and exocytosis.

Membrane transport: types. Diffusion- passive and facilitated. General classes of transport systems- uniport, symport, antiport. Active transport- primary and secondary. The P-type ATPases ( $\text{Na}^+\text{K}^+\text{-ATPase}$ ), F-type ATPases (ATP synthases), ABC transporters, ionophores, aquaporins, ion channels (ligand-gated and voltage-gated).

### **Unit-III Cell division, differentiation, cell cycle and cell death**

Molecular events in mitosis and meiosis. Brief account of cell differentiation.

The cell cycle: phases, regulation by cyclins and cyclin-dependent kinases. Checkpoints.

Cell death- types. Necrosis- causes and mechanism. Apoptosis: morphology, mitochondrial and death receptor pathways. Differences between apoptosis and necrosis.

### **Unit-IV Cell signaling**

Fundamental concepts and general features of cell signalling. Endocrine, paracrine, autocrine signaling and juxtacrine signalling. Types of receptors. Nuclear and cytosolic receptors. G-protein-coupled receptors. Second messengers: c-AMP, cGMP, diacylglycerol, inositol triphosphate and  $\text{Ca}^{2+}$ . Receptor tyrosine kinases- insulin signalling, ras-raf-MAP kinase and JAK-STAT pathways.

### **Unit-V Genetics**

Definitions of some common terms in genetics- phenotype, genotype, heterozygous, homozygous, allele (dominant, recessive, wild-type, mutant), character, gene, gene locus, pure line, hybrid. Mendel's laws. Monohybrid cross, multiple alleles, dihybrid cross, test cross, backcross, epistasis.

Chromosome structure. Polytene and lampbrush chromosomes. Types of chromosomes on the basis of centromere position. Karyotyping. Variation in chromosome number (euploidy, aneuploidy), arrangement (translocation, inversion), number of segments (deletion, duplication). Population genetics- The Hardy-Weinberg law (basic concept).

### **Text Books**

1. Karp. Cell & Molecular Biology 8<sup>th</sup> ed 2016. Wiley.
2. Lodish et al Molecular Cell Biology 8th ed. Freeman, 2017.
3. Nelson and Cox. Lehninger Principles of Biochemistry. Freeman, 7th ed. 2017.
4. Fletcher et al. Instant Notes in Genetics. 4<sup>th</sup> ed. Taylor & Francis. 2012.
5. Abouelmagd and Ageely. Basic Genetics. Textbook and Activities. Universal Publ. 2nd ed. 2013.
6. Elrod and Stansfield Schaum's Outline of Genetics. 5<sup>th</sup> ed. McGraw Hill. 2010.

#### Reference Books

Alberts et al Molecular biology of the Cell. 6th ed. Garland Sci. 2014.

#### BITP 103: PRACTICAL-I Biochemistry, Cell Biology & Genetics

1. Quantitative estimation of amino acids by ninhydrin method.
2. Estimation of proteins by Lowry *et al* method.
3. Thermal denaturation of DNA
4. Estimation of DNA by diphenylamine method.
5. Estimation of RNA by orcinol method.
6. Microscopic examination of epithelial cells, plant cells.
7. Isolation of lymphocytes.
8. Microscopic examination of blood cells, epithelial cells, plant cells, tissue types.
9. Karyotype analysis: Onion and human.
10. Buccal smear– Barr bodies.
11. Distinguishing mutant phenotypes of *Drosophila melanogaster* - *cry<sup>b</sup> mutants*.

#### Unit–III Manipulation of animal reproduction & characterization of animal genes

Manipulation of reproduction in animals. Artificial insemination, embryo transfer, *in vitro* fertilization. Embryo transfer in cattle and applications. Somatic cell cloning- cloning of Dolly. Ethical issues.

Production of recombinant vaccine for foot and mouth disease. Probiotics for disease control.

#### Unit–IV Gene transfer methods

Vectors for gene transfer in animals: adenovirus and retrovirus. Gene constructs- promoter/enhancer sequences for transgene expression in animals. Selectable markers for animal cells- thymidine kinase, dihydrofolate reductase, CAT. Transfection of animal cells- calcium phosphate coprecipitation, electroporation, lipofection, peptides, direct DNA transfer, viral vectors, microinjection.

#### Unit–V Transgenic animal technology

Methods for producing transgenic animals- retroviral, microinjection, engineered stem cell. Targeted gene transfer. Transgene integration and identification methods. Transgenic cattle, sheep, fish and pigs. Uses of transgenic animals. Transgenic animals as models of human disease. Ethical issues in transgenesis.

#### Text Books

1. Glick and Pasternak. Molecular Biotechnology. 4<sup>th</sup> ed. ASM Press 2009.
2. Primrose Twyman and Old. Principles of gene manipulation. 8th ed. Blackwell Sci 2016.
3. Watson et al. Recombinant DNA 3rd ed. Freeman. 2006.
4. Wilson and Walker. Principles and techniques of Biochemistry and Molecular Biology. 7th ed. Cambridge University Press 2012.
5. Singh B.D. Biotechnology. Expanding horizons. 2004 Kalyani Publ.

#### Reference Books

1. Freshney RI. Culture of animal cells: A manual of basic technique. 6<sup>th</sup> ed. Wiley-Liss, 2010.

#### BITP 305: PRACTICAL–IV (Analytical Techniques, Genetic Engineering, Plant and Animal Biotechnology)

1. Spectrophotometry: Analysis of standard curve, absorption spectrum of oxidized and reduced forms (NAD, NADH).
2. Separation of lipids by thin layer chromatography.
3. Separation of proteins by gel filtration - Demonstration.

Modification of seed protein quality. Suppression of endogenous genes by antisense (delayed ripening). Cytoplasmic male sterility.

### **Unit-V Transgenic plant technology-II**

Genetic modification of flower pigmentation. Terminator technology. Production of vaccines by transgenic plants. Problems in gene transfer in plants. Ethics of genetically engineered crops. Biotechnology and Intellectual Property Rights (IPR)-patents, trade secrets, copyright, trademark, TRIPS.

#### **Text Books**

1. Smith RH. Plant tissue culture. 3<sup>rd</sup> ed. Elsevier. 2013.
2. Primrose Twyman and Old. Principles of gene manipulation and Genomics. 8<sup>th</sup> ed. Blackwell Sci 2016.
3. Glick and Pasternak. Molecular Biotechnology. 4<sup>th</sup> ed. ASM Press 2009.
4. Watson et al. Recombinant DNA: Genes and genomes- A short course. 3<sup>rd</sup> ed. Freeman 2006.

#### **Reference Books**

Slater A. Plant Biotechnology: The Genetic Manipulation of Plants. 2<sup>nd</sup> ed., Oxford Univ Press, 2008.

## **BITC 304: ANIMAL BIOTECHNOLOGY**

**Objective:** To gain an insight into animal tissue culture techniques, gene transfer and gene manipulation methods, and transgenic animal technology.

### **Unit-I Animal cell culture-I**

Animal cell and tissue culture- merits and demerits. Laboratory facilities, substrate, culture media. Culture procedures- preparation, sterilization, disaggregation of tissue (mechanical and enzymatic), subculture, contamination. Primary culture, Secondary culture, cloning of cell lines, cancer cell lines. Measurement of cell viability and cytotoxicity. Large-scale culture of cell lines- monolayer, suspension and immobilized cultures.

### **Unit-II Animal cell culture-II**

Organ and histotypic culture- technique, advantages, limitations, applications. 3D cultures. Whole embryo culture. Somatic cell hybridization. Stem cells: types (embryonic, adult), isolation, identification, expansion, differentiation and uses, stem cell engineering, ethical issues. Commercial applications of animal tissue culture. Hazards and safety aspects of tissue culture.

## **BITC 201: ENZYME TECHNOLOGY**

**Objective:** To understand the basic aspects of enzyme action, kinetics, inhibition, and the applications of enzymes in industry.

### **Unit-I**

Enzyme- classification and nomenclature. Methods of enzyme isolation, purification and assay. Enzyme units: IU, Katal and Specific activity. Active site. Coenzymes, multienzyme complexes, metal-dependent and metalloenzymes. Isoenzymes. Extremozymes. Elementary details of ribozymes, abzymes and DNA enzymes.

### **Unit-II**

Enzyme kinetics: Effect of pH, temperature, enzyme and substrate concentration on enzyme activity. Michaelis-Menten plot, Lineweaver-Burk plot. Significance of  $K_m$  and  $V_{max}$ . Turnover number. Kinetics of allosteric enzymes- MWC and KNF models. Sequential and nonsequential bisubstrate reactions. Pre-steady state and steady state kinetics.

### **Unit-III**

Enzyme inhibition- irreversible and reversible competitive, noncompetitive, uncompetitive, mixed inhibition (derivation of rate equation not required). Mechanism of enzyme action- acid-base catalysis, covalent catalysis, strain, proximity and orientation effects. Mechanism of action of chymotrypsin. Enzyme regulation- zymogen activation, feedback inhibition. Covalent modification of enzymes and compartmentation. Allosteric regulation. Enzyme repression and induction.

### **Unit-IV**

Enzyme reactors: types (stirred tank, continuous flow), Immobilization of enzymes: principles, parameters, carriers, methods of immobilization and applications. Enzyme engineering principles and applications. Enzyme production and purification: enzyme sources, processes to improve enzyme yield, downstream processing of enzymes and chromatographic purification (brief account). Enzyme electrodes. Biosensors: components, types and applications.

### **Unit-V**

Use of enzymes in detergents, textiles, and leather industry, production of glucose syrup and cheese. Synzymes and solvent engineering. Soluble enzymes- applications in food, starch

processing and detergents. Elementary details of enzymes as diagnostic aids. Therapeutic uses of enzymes: enzymes as thrombolytic agents and digestive aids. Regulations and safety criteria for enzyme production and use.

#### Text Books

1. Palmer T. Understanding enzymes. Prentice Hall. 2004.
2. Buchholz et al Biocatalysts and Enzyme Technology. 2<sup>nd</sup>ed Wiley-Blackwell. 2012.
3. Pandey et al. Enzyme Technology. 2010, Springer.
4. Nelson, Cox. Lehninger Biochemistry. 7<sup>th</sup> ed. Freeman 2017.
5. Balasubramanian et al. Concepts in Biotechnology. Univ Press 2004.

#### Reference Books

Dixon and Webb. Enzymes 3rd ed. Longmans 1979.

### BITC 202: IMMUNOLOGY AND IMMUNOTECHNOLOGY

**Objective:** To acquire knowledge on immunological mechanisms and immunotechniques.

#### Unit-I

Types of immunity- innate and acquired. Humoral and cell mediated immunity. Central and peripheral lymphoid organs. Cells of the immune system- lymphocytes, mononuclear phagocytes-dendritic cells, granulocytes. NK cells, mast cells, interleukins.

Antigens-definition, antigenicity and immunogenicity, antigenic determinants, epitopes, haptens. Immunoglobulins-structure, classification and functions.

#### Unit-II

T-cell, B-cell receptors. Antigen recognition - processing and presentation to T-cells. Interaction of T and B cells. Immunological memory. Effector mechanisms- macrophage activation. Cell mediated cytotoxicity, immunotolerance, immunosuppression. Complement activation. Clonal selection theory. Immunoglobulin rearrangements, class switching.

#### Unit-III

Transplantation types. MHC antigens in transplantation. Immunodeficiency disorders-AIDS: The HIV genome and life cycle. Autoimmunity and elementary details of autoimmune disorders (systemic lupus erythematosus, rheumatoid arthritis). Hypersensitivity - types (basic concepts only).

#### Reference Books

1. Winnacker EL. From Genes to clones. 4<sup>th</sup> ed VCH Publ. 2003.
2. Watson et al. Recombinant DNA 3<sup>rd</sup> ed. 2006, Freeman.
3. Primrose, Twyman and Old. Principles of gene manipulation. 8<sup>th</sup> ed. Wiley-Blackwell. 2016.

### BITC 303: PLANT BIOTECHNOLOGY

**Objective:** To acquire theoretical knowledge about plant tissue culture techniques, gene transfer methods, and transgenic plant technology.

#### Unit-I Plant tissue culture-I

Totipotency of plant cells- dedifferentiation and redifferentiation. Sterilization. Nutrient medium. Use of growth regulators. Callus and suspension cultures. Single cells: isolation, culture techniques and applications. Organogenesis and somatic embryogenesis-techniques and applications. Anther, ovary, meristem culture. Somatic hybridization (Symmetric, Asymmetric, Cybrids). Embryo culture. Embryo rescue. Synseed production.

#### Unit-II Plant tissue culture-II

Large-scale culture of plant cells. Production of biochemicals from cultured plant cells. Micropropagation. Somaclonal and Gametoclonal variation. Cryopreservation and *ex situ* conservation of germplasm. Production of haploid plants and homozygous lines. Detection and uses of haploids in plant breeding. Protoplast isolation, culture and fusion, selection of hybrid cells and regeneration of hybrid plants.

#### Unit-III Cloning in plants

*A. tumefaciens* mediated transformation-Ti plasmids (cointegrate and binary vectors), direct nuclear transformation (protoplast transformation, particle bombardment), viral vectors (CaMV, TMV), chloroplast transformation. Use of reporter genes in transformed plant cells. Selectable markers for plants- drug resistance and herbicide resistance markers. RFLPs, RAPDs, DNA fingerprinting-general principles and applications in plant biotechnology.

#### Unit-IV Transgenic plant technology-I

Insect resistance plants- *cry* genes of *B.t.*, their proteins and target insects, *cry* gene expression in plants, insect resistance to *Cry* proteins. Strategies to obtain virus resistant transgenic plants. Herbicide resistance and stress - and senescence-tolerant plants.

### Unit–III Expression systems

Factors affecting expression of cloned genes. Expression of eukaryotic genes in bacteria- expression vector, promoters, industrial protein production. Fusion proteins, strategies to enhance protein stability, secretion and metabolic load. Expression in eukaryotic cells: Expression in yeast- yeast vectors, GAL system. Baculovirus and Mammalian expression systems (brief account). Tagged proteins and secretion signals. Reporter genes- types and uses.

### Unit–IV Techniques in Gene Manipulation

Extraction and purification of nucleic acids. Probes: radioactive and nonradioactive. Blotting techniques: Southern, northern, and western. Principle and applications of DNA fingerprinting, in situ hybridization, PCR, RT-PCR, real-time qPCR. DNA Sequencing: Automated sequencing. Next-generation sequencing. Site-directed mutagenesis (SDM): cassette and oligonucleotide-directed mutagenesis. PCR-based methods. Protein engineering by directed evolution and DNA shuffling. Hazards and safety aspects of genetic engineering.

### Unit–V Nanobiotechnology

Techniques for visualization of biomolecules at nanoscale- atomic force microscopy, optical microscopy, magnetic resonance force microscopy, TEM, SEM, FRET. Nanoparticles- metal, and bimetallic nanoparticles, quantum dots, dendrimers, and fluorescent nanoparticles. Production of nanoparticles: Collision/Coalescence mechanism of primary particle formation, nanoparticles agglomerates and aerogels. Biological synthesis of nanoparticles (brief account only). Applications of nanotechnology in biology, medicine and environment.

#### Text Books

1. Nicholls DTS. An Introduction to Genetic Engineering. 3rd ed. Cambridge Univ Press. 2008.
2. Glick and Pasternak. Molecular Biotechnology. 4<sup>th</sup> ed. ASM Press 2009.
3. Reece. Analysis of Genes and Genomes. Wiley 2004.
4. Jain KK. Nanobiotechnology Molecular Diagnostics: Current Techniques and Applications. Taylor & Francis. 2006.
5. vo-Dinh Nanotechnology in Biology and Medicine: Methods, devices and applications. CRC Press. 2<sup>nd</sup> ed. 2017.

### Unit–IV

Immunization practices-active and passive immunization. Vaccines-killed, attenuated-toxoids. Recombinant vector vaccines-DNA vaccines, synthetic peptide vaccines. Production and applications of polyclonal and monoclonal antibodies. Genetically engineered antibodies.

### Unit–V

Agglutination and precipitation techniques. Immuno-electrophoresis, RIA, Immunoblotting, Avidin-biotin mediated immunoassay. Immunohistochemistry, immune-fluorescence. Complement fixation test. ELISA-principle and applications. Flow cytometry.

#### Text Books

1. Goldsby *et al.* Kuby Immunology. WH Freeman & Co. 7<sup>th</sup> ed 2013.
2. Abbas *et al.* Cellular and Molecular Immunology. 9<sup>th</sup> ed. Elsevier 2018.
3. Janeway, C. (Ed), Immunobiology 9<sup>th</sup> ed. Garland Publ. 2016.
4. Coico and Sunshine. Immunology: A short course. 7<sup>th</sup> ed. Wiley, 2015.

#### Reference Books

- Roitt *et al.* Roitt's Essential Immunology. 13<sup>th</sup> ed Wiley-Blackwell Sci. 2017.

## BITC 203: INDUSTRIAL AND ENVIRONMENTAL BIOTECHNOLOGY

**Objective:** To learn the principles and techniques of bioprocess engineering and downstream processing as well as biotechnological approaches to environmental management.

### Unit–I Bioprocessing and bioreactors

Bioprocess engineering: Isolation and screening of industrially important microbes. Maintenance of strains. Inoculum development. Strain improvement- mutant selection, recombination, metabolite production by rDNA technology. Process development. Advantage of bioprocess over chemical process. Kinetics of microbial growth and death. Bioreactors- design, function and types. Media for industrial fermentation. Antifoaming devices. Analysis of batch, fed-batch and continuous bioreactions.

### Unit–II Downstream processing

Downstream processing: Stages: solid-liquid separation, release of intracellular compartments, concentration of biological products, membrane filtration, precipitation, adsorption, dialysis, reverse osmosis, ultrafiltration, preservation and stabilization, purification.

Crystallization and drying. Product formulation. Monitoring of downstream processing.

Industrial production of ethanol, citric acid, penicillin, and lysine/aspartate. Whole cell immobilization and industrial applications.

#### **Unit–III Pollution and control**

Environmental pollution- types, methods for measurement, biosensors to detect environmental pollutants, hazards from wastes and pollutants. Air pollution and its control through biotechnology. Water pollution and control. Wastewater treatment- physical, chemical and biological. Activated sludge- oxidation ditches and ponds, trickling filter, towers, rotating discs and drums. Anaerobic processes: anaerobic digestion and filters. Effluent treatment: B.O.D and C.O.D Treatment for wastewaters of distillery, dairy, and tannery industries.

#### **Unit–IV Soil and agricultural biotechnology**

Soil microbiota. Growth, ecological adaptations, interactions among soil microorganisms, biogeochemical role of soil microorganisms. Microbial degradation of xenobiotics in the environment. Oil spill clean up. Bioremediation of contaminated soil and waste land. Biofertilisers. Biopesticides in integrated pest management- *Bacillus* and baculoviruses as biocontrol agents. Biodegradable plastics. Biofilms.

#### **Unit–V Alternative energy sources and green technology**

Renewable sources of energy (solar, wind, biogas, energy crops, cellulose); hydrogen production using hydrogenase and nitrogenase. Conservation of energy. Bioleaching- use of microorganisms in mining of gold and uranium. Global environmental problems; Ozone depletion, greenhouse effect, impact and management. Reforestation through micropropagation- use of *Casuarina*, and mycorrhizae. Development of stress resistant plants. Biodiversity- Alpha and beta diversity. Extinction and endangered species. Conservation of biodiversity. *In situ* and *ex situ*- gene banks, species conservation.

#### **Text Books**

1. Ratledge and Kristiansen. Basic Biotechnology 3rd ed. Cambridge Univ. Press 2006.
2. Borem, Santos, Bowen. Understanding Biotechnology. Pearson, 2011.
3. Casida L.E. JR Industrial Microbiology. 3rd Wiley Eastern reprint. John Wiley and Sons Inc. 2<sup>nd</sup> ed. 2016.

#### **Unit–V Centrifugation**

Basic principles of sedimentation. Types of rotors. Low-speed and high-speed centrifuges. Analytical and preparative ultracentrifuge-instrumentation and applications. Subcellular fractionation by differential centrifugation. Density-gradient centrifugation- rate zonal and isopycnic.

#### **Text Books**

1. Wilson and Walker. Principles and techniques of Biochemistry and Molecular biology. 7th ed. Cambridge University Press 2012.
2. Upadhyay, Upadhyay and Nath. Biophysical Chemistry principles and Techniques. Himalaya Publ. 2010.
3. Boyer, R. Modern Experimental Biochemistry. 3rd ed. Addison Wesley Longman, 2000.

#### **Reference Books**

1. Sambrook. Molecular Cloning. Cold Spring Harbor Laboratory, 4<sup>th</sup> ed 2012.
2. Friefelder and Friefelder. Physical Biochemistry - Applications to Biochemistry and Molecular Biology. WH Freeman & Co. 1994.
3. Pavia Intro to spectroscopy, 5<sup>th</sup> ed., 2015.

### **BITC 302: GENETIC ENGINEERING AND NANOBIOTECHNOLOGY**

**Objective:** To master the basic principles of genetic engineering, cloning strategies, and techniques. To learn the basics and applications of nanobiotechnology

#### **Unit–I Restriction endonucleases, cloning vectors, and ligation**

Basic steps in gene cloning. Type II Restriction endonucleases- nomenclature and types of cleavage. Cloning vectors: plasmids (pBR322 and pUC), phage vectors ( $\lambda$ ), cosmids, BACs and YACs. Methods of ligation of insert and vector DNA molecules: cohesive end method, homopolymeric tailing, blunt-end ligation, linkers and adapters.

#### **Unit–II Gene transfer methods, cloning strategies & screening**

Gene transfer methods: calcium phosphate coprecipitation, electroporation, lipofection, viruses, microinjection. Choice of host organisms for cloning. Cloning strategies- genomic cloning, cDNA cloning. Differences between genomic and cDNA libraries. Screening of recombinants: marker inactivation (antibiotic resistance, blue-white selection), colony hybridization, immunoscreening, screening for protein activity.

15. Isolation and enumeration of soil bacteria.
16. Identification of heavy metals from sewage water by AAS (demonstration)

### **BITC 301: ANALYTICAL TECHNIQUES**

**Objective:** To learn the principle, operation, and applications of various techniques for analyzing biomolecules. This course will help students to understand the theoretical basis of separation and analysis of biomolecules which form the basic tools in research.

#### **Unit-I Spectroscopy techniques**

Laws of absorption. Absorption spectrum. Principle, instrumentation and applications of UV-visible spectrophotometry, spectrofluorimetry and luminometry. Atomic spectroscopy- principle and applications. Brief outline of the principles and biological applications of NMR and ESR, ORD, and CD.

#### **Unit-II Radioisotope Techniques and Microscopy**

Nature and units of radioactivity. Solid and liquid scintillation counting, quenching, scintillation cocktails and sample preparation. Autoradiography. Applications of radioisotopes in biology. Radiation hazards.

Microscopy- basic principles, and components of light, bright field, phase contrast, fluorescence and confocal microscopy. Electron microscopy- principle, preparation of specimens for TEM and SEM.

#### **Unit-III Electrophoresis**

Electrophoresis: General principles. Support media. Electrophoresis of proteins- SDS-PAGE, native gels, gradient gels, isoelectric focusing. Cellulose acetate electrophoresis. Electrophoresis of nucleic acids- agarose gel electrophoresis, pulsed field gel electrophoresis. Electrophoretic mobility shift assay. DNase I footprinting.

#### **Unit-IV Chromatography**

General principles of partition and adsorption chromatography. Principle, instrumentation and applications of thin layer and gas chromatography. Principle, procedure, and applications of ion-exchange, molecular exclusion, and affinity chromatography. HPLC- principle, instrumentation and applications.

4. Smith JE. Biotechnology Cambridge University Press. 5<sup>th</sup> ed., 2012.
5. Gupta PK. Elements of Biotechnology, Rastogi Publication, 2<sup>nd</sup> ed., 2016.
6. Scragg A. Environmental Microbiology 1st ed. Am Society for Microbiology 2005.
7. Ahmed N. Industrial and environmental Biotechnology. Horizon Scientific Press 2001.
8. Primrose Twyman and Old. Principles of gene manipulation. 8th ed. Blackwell Sci 2016.

#### **References**

Flickinger and Drew (eds). Encyclopedia of Bioprocess Technology. 5 vol. set. John Wiley & Sons, 1999.

### **BITC 204: MOLECULAR BIOLOGY**

**Objective:** To gain an insight into the molecular mechanisms of genetic information flow and regulation of gene expression.

#### **Unit-I Chromatin and Genome complexity**

The central dogma of molecular biology. The *E. coli* chromosome and DNA-binding proteins. Plasmids- classification and properties. Eukaryotic chromatin: nucleosomes, 30 nm fiber and higher order chromatin structure. Concept of the gene. Definitions of the following: gene, cistron, coding region (ORF), transcription unit, untranslated region (UTR), pseudogenes, euchromatin and heterochromatin. Typical structure of protein-coding genes in prokaryotes and eukaryotes. Split genes- exons and introns. DNA sequence elements: unique sequence DNA, repetitive DNA (SINEs, LINEs, satellite, minisatellites and microsatellites).

#### **Unit-II Replication, Repair and Recombination**

Messelson and Stahl experiment. Enzymes and proteins involved in replication: helicases, SSB, topoisomerases, DNA polymerases, DNA ligase. DNA replication in bacteria and eukaryotes: initiation, elongation, termination. The end-replication problem and telomerase. Inhibitors of replication.

DNA damage by physical and chemical agents. DNA repair- photoreactivation, excision repair, mismatch repair, SOS response, double strand break repair. Molecular biology of homologous recombination. Transposons: mechanism of transposition and applications.

#### **Unit-III Transcription and Post-transcriptional processing**

Transcription in *E. coli*: RNA polymerase subunit structure, promoter sequence steps in transcription- template recognition, initiation, elongation and termination (intrinsic, rho-dependent). Transcription in

eukaryotes: RNA pol I, II and III: subunit structure, transcription factors, promoters, inhibitors. Mechanism of RNA pol II transcription: preinitiation complex formation, transcription initiation (activator proteins, mediator, chromatin recruitment), elongation, termination.

Classes of introns. Post-transcriptional processing of prokaryotic and eukaryotic rRNA, and tRNA. and eukaryotic mRNA. Brief account of ribozymes, RNA editing and Reverse transcription.

#### **Unit IV Genetic code and Translation**

The genetic code: general features. Mitochondrial genetic code. Mutations: point mutations and frameshift mutations. Suppressor mutations- nonsense and missense suppression.

Mechanism of protein synthesis in bacteria and eukaryotes: amino acid activation, initiation, elongation and termination. Inhibitors of protein synthesis. Post-translational modifications. Protein targeting to subcellular organelles, secretory proteins (the signal sequence hypothesis). Protein degradation: the ubiquitin pathway. Protein folding- models, molecular chaperones.

#### **Unit-V Regulation of Gene expression**

Basic principles of gene regulation- levels of gene expression, definition of housekeeping genes, and inducible genes, upregulation, downregulation. Regulation of gene expression in prokaryotes: *lac* operon and *trp* operon. Translational control in bacteria (r-protein operons). Regulation of gene expression in eukaryotes: Transcriptional regulation by steroid hormone receptors, phosphorylation (STAT proteins), alternative splicing. Translational regulation. Antisense RNA and RNA interference. Epigenetic gene regulation: DNA methylation, histone acetylation and deacetylation.

#### **Text Books**

1. Nelson and Cox. Lehninger Principles of Biochemistry. Freeman, 7th ed. 2017.
2. Krebs JE et al. Lewin's. Genes XII. Jones & Bartlett Publ, 2017.
3. Alberts et al Molecular biology of the cell. 6th ed. Garland Sci. 2014.
4. Watson. Molecular Biology of the Gene. 7th ed. Pearson Edu, 2013.

#### **Reference Books**

1. Watson et al. Recombinant DNA: Genes and genomes - A short course. 3<sup>rd</sup> ed. Freeman 2006.
2. Twyman. Advanced Molecular Biology. BIOS Sci Publ. 2000.

#### **BITP 205: PRACTICALS IN ENZYME TECHNOLOGY, IMMUNOTECHNOLOGY AND MOLECULAR BIOLOGY**

1. Determination of total and specific activity of  $\alpha$ -amylase.
2. Effect of pH on enzyme activity (amylase).
3. Effect of temperature on enzyme activity (amylase).
4. Effect of substrate concentration on enzyme activity (amylase) and determination of  $K_m$  value.
5. Identification of blood groups and Rh typing.
6. Radial immunodiffusion.
7. Double diffusion.
8. Immunoelectrophoresis.
9. Preparation of antisera.
10. Isolation of DNA, DNA electrophoresis in agarose gel, determination of molecular weight.
11. Isolation of RNA from yeast.
12. Bacterial conjugation, transformation (Demonstration).

#### **BITP206:PRACTICAL-III (Microbiology, Industrial and Environmental Biotechnology)**

1. Microscopic examination of bacteria, fungi, yeast.
2. Staining of microorganisms: Gram staining, acid fast staining.
3. Culture of microorganisms: media preparation, Serial dilution, inoculation, Culture of bacteria in culture tubes, agar plates.
4. Antibiotic sensitivity and resistance pattern of bacteria.
5. Preparation of immobilized cell/enzyme beads using alginate.
6. Determination of growth curve of a microorganism and substrate degradation profile.
7. Comparative studies of ethanol production with different substrates.
8. Microbial production of citric acid using *Aspergillus niger*.
9. Determination of total dissolved solids in water.
10. Determination of D.O. concentration of water sample.
11. Determination B.O.D. of sewage sample.
12. Determination C.O.D. of sewage sample.
13. Estimation of nitrate in drinking water.
14. Efficiency of removal of air pollutants using fibrous air filter.