Enclosure - II

SEMESTER III

16PMBCC09	Core 7: Gene Manipulation Techniques	4hrs/week	4Credits

Objectives

After completion of this course, student will be able to:

- Understand the basics of Enzymes and Vectors used in Gene manipulation
- Describe the methods used in selection, screening & analysis of recombinants
- Develop knowledge of the genomic and cDNA cloning strategies
- Understand the strategies used in genes cloning

UNIT-1: Enzymes involved in Genetic Engineering (10 hrs)

- Endo and Exo Nuclease: Classification, Mechanisms of enzyme action and role in Genetic engineering
- Restriction Endonuclease:Classification, Mechanisms of enzyme action and role in Genetic engineering
- Ligases –Classification, Mechanism of enzyme action and role in Genetic engineering
- Additional enzymes – DNA Polymerase, RNA Polymerase, Alkaline Phosphatases, Reverse transcriptase, Polynucleotide phosphorylase, polynucleotide kinases.

UNIT-2: Gene cloning vectors and tools (10 hrs)

- Plasmid: Basic biology, Natural and Synthetic plasmid, Role of Plasmid in Genetic engineering.
- Phages: Basic biology and Role of Bacteriophage in Genetic engineering. Example of bacteriophage vector - Lambda phage vectors
- Artificial vectors : Cosmids, Phagemids, BAC, YAC, Shuttle vector
- Expression Vector – Vectors to facilitate protein purification, promote solubilization of expressed proteins
- Adaptors, Linkers, Homopolymer tailing

UNIT-3: Cloning Strategies **(10 hrs)**

- Genomic libraries, PCR as an alternative to genomic DNA cloning
- c-DNA Synthesis & cloning, Full-length cDNA cloning
- Rapid amplification of cDNA ends (RACE)
- Probe preparation (Radiolabelled & non-radiolabelled)

UNIT-4: Screening, Selection & Analysis of recombinants **(8 hrs)**

- Basic techniques for screening and selection of the clones:- use of chromatography substrate, Insertional inactivation, Complementation of defined mutation
- Sequence-dependent screening: Screening by hybridization, Screening by PCR
- Screening expression libraries : Immunological screening, South-Western and North-Western screening
- Functional cloning: Functional complementation, Screening by 'gain of function', Differential screening.
- Positional cloning, Chromosome walking and jumping

UNIT-5: Advanced Techniques **(10 hrs)**

- PCR: Introduction, Types and Applications
- Sequencing of Nucleic acid : Enzymatic DNA sequencing, Chemical sequencing of DNA,Shotgun sequencing and Next-generation methods
- DNA markers:- RFLP, micro-minisatellites, SNP's, RAPD's, AFLP
- Application of Genetic engineering:
  - ✓ Transgenic plants: Bt cotton, roundup ready soybean
  - ✓ Production of edible vaccines and biotech drugs.

Suggested Reading:

1. S.B. Primrose, R.M. Twyman and R.W.Old.(2001) *Principles of Gene Manipulation. 6th Edition*, S.B.University Press,.

2. J. Sambrook and D.W. Russel.(2001) *Molecular Cloning: A Laboratory Manual, Volume 1-3*, CSHL,

3. Brown TA.(2006).*Gene Cloning, 3rd ed*. Garland Science

16PMBCC10	Core 8:Immunology and Medical Microbiology	4hrs/week	4Credits

Objectives:

After completion of this course, student will be able to :

1. Define and describe the cells and organs of immune system and distinguish between innate and adaptive immune responses.
2. Understand the structure and function of antigens and antibodies, basis of their interaction and carry out diagnostic tests based on these interactions
3. Comprehend the molecular basis of transplantation immunology and predict its success in different scenarios
4. List out medically important microbes, understand mechanism of their pathogenesis and decide their prevention and control strategies

Unit 1: Basics of Immunology (8 hrs)

- Introduction and scope of Immunology
- Immunity : Types of immunity (Innate, Acquired, Active and Passive)
- Antigens : Characteristics types, epitopes factors affecting antigenicity
- Antibody : Basic Structures, functions, classes, immunoglobulin variants, mechanism of antibody formation, monoclonal & Polyclonal antibody
- Antigen antibody interactions

Unit 2: Immune system (9 hrs)

- Complement system: Classical and Alternative pathways and its regulations
- Cells of immune systems : Heamatopoiesis, Lymphocytes, Monocytes, Granulocytes Dendritic cells and Mast cells
- Organs of immune systems : Primary & Secondary lymphoid organs
- Immune response : Cell mediated, antibody mediated, MHC, immunological memory, Immunological tolerance
- Hypersensitivity and Immunohaematology

Unit 3: Transplantation, Tumor Immunology & Vaccines (9 hrs)

- Immunology of tissue transplantation : Transplantation immunology, graft acceptance and rejection, clinical manifestation and prevention of graft rejection
- Tumor Immunology : Basics of cancer, Tumor antigens, Tumor evasion of immune system, tumor immunotherapy
- Immunodeficiency diseases - Primary Immunodeficiency (AIDS) and Secondary Immunodeficiency (SCID)
- Autoimmunity and autoimmune diseases - Organ specific (Graves disease, Insulin dependent diabetes mellitus) Systemic Autoimmune Diseases (Rheumatoid Arthritis, Multiple sclerosis).

Unit 4: Medically important organisms (12 hrs)

- Identification, pathogenicity, diagnosis and prevention/control of disease caused by the following organisms:
  - Bacteria : *Staphylococci, Streptococci, Pneumococci, Neisseriae (Gonococci & meningococci), Corynebacterium, Mycobacterium, Clostridium, Bacillus, Salmonella, Shigella, Vibrio, Enterobacter*
  - Viruses : *Pox virus, Herpes virus (HSV I & II) Adenovirus, Picorna virus, Orthomyxoviruses, (influenza), Paramyxoviruses (Mumps and Measles), Rhabdoviruses, Hepatitis viruses, HIV, DENV, Ebola virus*
  - Fungus : *Blastomyces, Coccodiodomyces, Candida, Opportunistic mycoses Aspergillus*
  - Protozoa : Protozoan diseases - Malaria, Leishmaniasis and Filariasis.

Unit 5: Disease management and Control (10 hrs)

- Epidemiology and disease cycle of Dengue fever, Malaria
- Molecular pathogenesis and virulence factors of *S. aureus*
- Nosocomial infection : Occurrence & Drug resistance
- Host Parasite interactions in animals
- Zoonotic diseases and their control

Suggested Readings

1. R. Ananthanarayanan and CK JayaramPanicker (1997) Text of Microbiology, Orient Longman.
2. Mackie and McCartney (1996) Medical Microbiology, Practical Medical Microbiology Churchill Livingstone.
3. Shanson, D.C., Wright, P.S.G, (1982) Microbiology in Clinical Practice.
4. Finegold, S.M., Baron, E.J. Bailey, W.R (1990) Bailey and Scott's Diagnostic Microbiology 8th Edition, Published by Mosby, St. Louis, MO.
5. Smith, C.G.C. (1976): Epidemiology and Infections. Medowfief Press Ltd., Shildon, England.
6. Kindt T.J., Osborne, B.A., Goldsby, R. (1994) *Kuby Immunology*6th Edition W.H. Freeman and Co. New York.
7. Male, D., Brostoff, J., Roth, D.B., Roitt I.V. (2012) *Immunology*. Elsevier Publication.
8. Khan, F.H. (2009) *The elements of Immunology*Pearson Education India.
9. Rose, N.R.(2002) *Manual of Clinical Laboratory and Immunology* 6th Edition. ASM Publications.
10. Owen, J.A., Kuby J, Punt, J., Stranford, S.A. (2013) Kuby Immunology 7th Edition Macmillan Publication.
11. Dubey, R. C. (1993). A textbook of Biotechnology. S. Chand Publishing.

16PMBCC11	Core 9:Basic Instrumentation and Biophysics	4hrs/week	4Credits

Objectives:

1. To understand the basic principles of biophysical instrumentation applied in diverse field of Microbiology
2. To understand the way of studying the biological molecules using analytical techniques
3. To study the application of the biophysical instruments

Unit 1: Microscopy and Radio isotopic techniques

- Light Microscopy: - Bright field, Dark field, Fluorescent microscopy, Phase contrast microscopy
- Electron Microscopy (TEM & SEM), Atomic Force Microscopy
- Flow cytometry (FACS)
- Radioisotopes & half life of isotopes, Units & measurement of radiation, Autoradiography, Application of radioisotopes in biological study, Interaction of radiation with matter

Unit 2: Spectroscopy

- Spectroscopic techniques:- Beer Lambert's law, Extinction coefficient, Principles & applications of visible & U.V. spectroscopic technique
- Electromagnetic spectrum, interaction of electromagnetic radiation with matter, Physical phenomenon:- Absorption, Emission, Refraction, Diffraction, Transmission
- Absorption & Emission Spectroscopy
- CD spectroscopy, Raman's Spectroscopy, IR & NMR
- X-ray diffraction & crystallization

Unit 3: Chromatography techniques

- Chromatography : Theory & Principles
- Understating of basic terminology: Stationary phase, mobile phase, Retention time, column efficiency, Peak shape
- Types of chromatography, partition, adsorption, ion exchange, size exclusion, affinity, Paper chromatography,

- Hydrophobic chromatography, Gas chromatography, Ultra high performance chromatography, Liquid chromatography combine with Mass spectroscopy, Thin layer chromatography, High performance thin layer chromatography

Unit 4: Centrifugation & Electrophoresis

- Basics, principles and classification of electrophoresis
- Zone Electrophoresis : Paper electrophoresis, Thin layer electrophoresis, Cellulose acetate electrophoresis, gel electrophoresis, affinity electrophoresis
- Moving boundary electrophoresis: Capillary electrophoresis, Immuno electrophoresis
- Basics, principles and classification centrifugation
- Types Centrifugation, Sedimentation, Relative centrifugal force, preparative and analytical centrifuge, ultracentrifugation and its applications in molecular size determination.

Unit 5: Advanced Biophysics

- Aspects of advanced biophysics : Concepts, principles and applications.
- Electrophysical techniques in diagnostics: Single neuron recording, patch-clamp recording, electrocardiogram, Brain activity recording, lesion and stimulation of brain, PET, MRI, fMRI, CAT, Density.
- CT Scanners and Their Applications, Overview of Digital Subtraction Radiography and Mammography
- Role and applications of biophysics in nuclear medicines, Principle of localization & usages of radiopharmaceuticals
- Practical aspects of Implementation of Radiation Protection in Medical Applications, Regulatory Aspects of Radiation Protection.

Reference Books:

1. Sambrook, J., Fritsch, E. F., &Maniatis, T. (1989). *Molecular cloning* (Vol. 2, pp. 14-9). New York: Cold spring harbor laboratory press.
2. Blau, K., & King, G. S. (Eds.). (1993). *Handbook of derivatives for chromatography* (Vol. 2). New York: Wiley.
3. Kindt, T. J., Goldsby, R. A., Osborne, B. A., &Kuby, J. (2007). *Kuby immunology*.

Macmillan.

4. Hayat, M. A. (1974). *Principles and techniques of scanning electron microscopy. Biological applications. Volume 1*. Van Nostrand Reinhold Company.
5. E Alpen (1997) *Radiation Biophysics,* 2nd Edition academic press
6. R.N. Roy. (2001) A *Textbook of Biophysics*. New Central Book Agency

16PMBDC10/ 16PBTDC10	DSEIII: Pharmaceutical Technology	4hrs/week	4Credits

Objectives:

1. For better perception in pharmaceutical technology
2. To understand industrial requirement of microbial technology
3. To get better opportunities in pharmaceutical industry/laboratories/ Research institutes
4. To be equipped with standard operating procedures as per regulatory authorities

Unit 1: Introduction to pharmaceutical industry (9hrs)

- Role of a Microbiologist/ Biotechnologist in pharma industry
- Active Pharmaceutical Ingredients units, Formulation units, Research and Development units,
- Quality Assurance and regulatory aspects.
- Pharmacopoeias with special reference to Indian, British, United States.
- Government regulatory practices and policies, FDA perspective.

Unit 2: Quality Assurance and Practices in Pharmaceutical Industry (10hrs)

- (GMP) and Good Laboratory Practices (GLP) in pharmaceutical industry.
- Concepts of pharmaceuticals, biologics and biopharmaceuticals, sources of biopharmaceuticals and its production
- Design and layout of sterile product manufacturing unit (Designing of microbiology laboratory).
- Health safety & environment (HSE), Safety in microbiology laboratory.
- Quality assurance and quality management in pharmaceuticals : ISO, WHO and US certification. CFR's, Compliance, CAPA and Deviations.

Unit 3: Quality analysis in Pharmaceutical Industries (10 hrs)

- Standard operating procedures for assay of antibiotics, vitamins and amino acids

- Antimicrobial testing of pharmaceutical products. Microbial Examination of Sterile & Non-Sterile Products
- Microbial limit test of pharma products (MLT),Sterility testing, Investigating USP Sterility Testing Failure, Water analysis
- Bacterial Endotoxin Testing (BET), Particulate Matter, Bioburden Estimation for Medical Devices
- Environmental Monitoring, Sterilization-heat, D-value, Z-value and survival curve, radioactive, gaseous and filtration. Chemical and biological indicators.Growth promotion test, Calibration and validation of equipments.

Unit 4: Antimicrobial resistance and drug delivery system (10hrs)

- Bacterial resistance to antibiotics: Origin, mechanism, transfer, and clinical implications.
- Molecular principles of drug targeting
- Microbial contamination and spoilage of pharmaceutical products (Sterile injectables, non sterile products, ophthalmic preparations and implants) and their sterilization.
- Chemical disinfectants, antiseptics and preservatives.
- Drug delivery system in gene therapy, Microencapsulation, Nanoparticles, Liposomes, Antibodies for drug delivery and Penetrating defenses.

Unit 5: Advances in Pharmaceutical Technology (9 hrs)

- Production of biopharmaceuticals by genetically engineered cells
- Advance techniques in manufacturing products
- Hormones (Humulin, Humatrope), Interferons (Intron A, Referon-A), t- Plasminogen activator
- New vaccine technology: DNA vaccines, synthetic peptide vaccines, multivalent subunit vaccines and Vaccine clinical trials.
- Application of microbial enzymes in pharmaceutical industry.

Suggested Readings

1. Stephen P. D., Norman A. H., Sean P. G., Brendan F. G. (2011)*Hugo & Russell Pharmaceutical Microbiology* 8th Ed. Wiley-Blackwell Publishing house
2. John S. Wolfson and David C. Hooper, (1989*) Quinolone antimicrobial agents*. American Society for Microbiology, Washington.
3. Cooper M. S. (1972) *Quality control in the Pharmaceutical Industry* Vol.2 Academic Press Inc.
4. Vyas S. P., Dixit V. (2007) *Pharmaceutical Biotechnology,* CBS Publishers & Distributors
5. Sidney H.W. Murray M. Tuckerman, W., S.Hitchings IV. Mercel D.,(2007) *Good Manufacturing Practices for Pharmaceuticals,* Second Edition, NC New York

16PMBCC13	Combined Practical Core - III Genetic Engineering, Immunology & Instrumentation	6hrs/week	3Credits

1. Restriction Digestion of λ DNA using three Restriction Endonuclease enzymes:
    a. a) EcoR V b) Hind III c) BamH I
2. Determination of Tm values of DNA
3. Plasmid Curing by Acridine Orange
4. Isolation & Characterization of plasmid DNA
5. Cloning in Plasmid or Phage vectors
6. Total & Differential Count of blood cells
7. Gel Techniques; SDS PAGE/Western blot
8. Analysis of compound by U.V. spectroscopic technique
9. Analysis of compound by IR spectroscopic technique
10. Purification of Immunoglobulin
11. Isolation and identification of clinically important microbes from clinical specimens(throat swab, sputum, nasal swab, urine, blood, stool)
12. Identification of pathogens on selective, differential and enrichment media
13. Different staining techniques a) Ziehl-Neelsen method of AFB b) Fluorochrome staining c) Leishman's staining d) Giemsa's staining
14. Grams Staining and Special staining methods to demonstrate granules, capsules and spores
15. Testing of drug susceptibility according to NCCLS
16. Determination of MIC by Kirby-Bauer method.
17. Induction and purification of antibodies
18. Precipitation reaction and Agglutinations (slide)
19. Blood grouping and Rh typing

16PMBDC13/ 16PBTDC13	DSE – III Practical (Pharmaceutical Technology)	2hrs/week	1Credits

Practicals:

1. Sterility testing of pharmaceutical products
2. Microscopic analysis of sterile injectables and tablets
3. Bacterial Endotoxin Test of pharmaceutical products
4. Microbiological assay of antibiotics in market
5. Microbial limit test (MLT) of water & pharmaceutical products
6. Isolation and identification microorganisms from syrups and sugary formulations
7. Bioburden estimation for sterile medical devices and equipments available in market

SEMESTER IV

16PMBCC14	Core 11:Bioinformatics	4hrs/week	4Credits

Objectives:

Upon completion of this course students will be able to

- Browse, search, and retrieve biological data from public repositories & describe the contents and properties
- Upload new sequences onto GenBank
- perform text- and sequence-based searches, and analyze the results
- explain the major steps in pairwise and multiple sequence alignment, explain the principle for, and execute pairwise sequence alignment by dynamic programming
- Edit raw Sanger sequence data for phylogenetic analysis (edit chromatograms, identify
- Contamination, align sequences, remove ambiguously aligned sites)
- Obtain basal knowledge of phylogentic theory as well as various analytic tools which will enable them to analyze different kind of data and interpret the result.
- Explain the major features of methods for modelling protein structures and use programs for visualizing and analyzing such structures.

Unit 1: Introduction and Bioinformatics Resources (10 hrs)

- Introduction to Bioinformatics: Definition, role, scope in different areas and current perspective.
- Database concepts, Biological Databases
- Nucleic acid sequence database: GenBank, ENA, DDBJ.
- Protein Resources: UniProtKB, SWISS-PROT, TrEMBL,
- Secondary sequence databases: PROSITE, Pfam, PRODOM.

Unit: 2 Databases & sequence alignments (10 hrs)

- Structure database: PDB, NDB
- Small Molecule database: Drug bank, Pubchem, ZINC
- Basic concepts of sequence alignment
- Needleman & Wunsch, Smith & Waterman algorithms for pair wise alignments,

- Multiple sequence alignment : Concept, Algorithm, tools and importance

Unit 3: Sequence analyses & primer designing (9hrs)

- Biological sequences file formats: genbank, fasta, gcg, msf, nbrf-pir etc.
- Sequence similarity: similarity, identity and homology
- Scoring matrices: basic concept of a scoring matrix, PAM and BLOSUM series.
- Sequence-based database searches: BLAST and FASTA
- Primer designing

Unit 4: Phylogenetic Analysis (9hrs)

- Phylogenetic analysis: Description and types of trees
- Computational models in phylogenetics: Various computational models of phylogenetic and molecular evolutionary analysis.
- Tree construction methods: Distance based
- Maximum Parsimony and Maximum Likelihood
- Tree Evaluation: Bootstrap and its computational aspects

Unit 5: Structural Bioinformatics and Drug designing (10hrs)

- Structural Bioinformatics: Introduction, coordinate systems, Visualization & presentation of structure.
- Secondary structure: algorithms of Chou Fasman, GOR methods.
- Tertiary Structure: Homology modeling, threading method.
- Protein structure alignment & structure assessment methods
- Introduction to drug discovery: History, analogue and structural drug discovery, ligand designing and optimization, Molecular docking – concept and methods.

Suggested Reading:

1. Lesk, A. (2013). Introduction to bioinformatics. Oxford University Press.
2. Mount, D. W., & Mount, D. W. (2001). Bioinformatics: sequence and genome analysis (Vol. 2). New York:: Cold spring harbor laboratory press.
3. Rastogi, S. C., Mendriratta, N., &Rastogi, P. (2006). Bioinformatics: Concepts, Skills & Applications. CBS Publishers & Distributors Pvt. Limited.
4. Baxevanis Andreas, D., Davison Daniel, B., Page Roderic, D. M., Petsko Gregory, A., Stein Lincoln, D., &Stormo Gary, D. (2003). Current protocols in bioinformatics.
5. Higgens, D. G., Taylor, W. R., & Webster, D. M. (2000). Protein Structure Prediction: Methods and Protocols.
6. Rastogi, S. C., Rastogi, P., &Mendiratta, N. (2008). Bioinformatics Methods And Applications: Genomics Proteomics And Drug Discovery 3Rd Ed. PHI Learning Pvt. Ltd.
7. Xiong, J. (2006). Essential bioinformatics. Cambridge University Press.
8. Baxevanis, A. D., & Ouellette, B. F. (2004). Bioinformatics: a practical guide to the analysis of genes and proteins (Vol. 43). John Wiley & Sons.
9. Eidhammer, I., Jonassen, I., & Taylor, W. R. (2004). Protein Bioinformatics: An algorithmic approach to sequence and structure analysis (Vol. 1). Chichester: Wiley

16PMBDC17/ 16PBTDC17	DSE IV: Environmental Biotechnology	5hrs/week	5Credits

Course specific outcomes

After completion of this course, student will be able to:

- Define and describe various methods of solid and liquid waste management.
- Comprehend the use of microbes in biominerelization and biohydrometallurgy for extraction of metals from ores.
- Understand the principles and process of removal of toxic substances and degradation of xenobiotics using microbes.
- Understand the role of Biotechnology in solving global environmental problems.

Unit1: Basics of Environmental Biotechnology (8 hrs)

- Introduction to Environmental Biotechnology
- Global environmental problems: Ozone depletion, Greenhouse effect and Acid rain
- Biodeterioration
- Eutrophication and Biomagnifications
- Toxic chemicals in the environment and their effects - air, water & soil

Unit 2: Waste Management (10hrs)

- Solid waste - Sources, generation and classification
- Management methods of solid- Sanitary land filling, Recycling, Composting and Incineration
- Liquid waste – Sources and types of liquid waste
- Treatment schemes for waste waters- Aerobic processes: Activated sludge, Oxidation ditches, Trickling filter, Towers, Rotating discs, Rotating drums
- Anaerobic digestion

Unit 3: Biodegradation and Bioremediation (10hrs)

- Bioremediation- processes/strategies and organisms involved
- Bioremediation: Degradation of pesticides & Xenobiotics
- Degradation of Basic Structures found in Hydrocarbons & Oil spills
- Overview of Phytoremediation
- Biomining : Use of microbes in biohydrometallurgy and biomineralization

Unit 4: Biosensors and Biofuels (10hrs)

- Application of microbes as Biofertilizers
- Bioinsecticides for productivity improvement and crop protection
- Principles of Biomonitoring
- Applications of Biosensors for detection of environmental pollutants
- Biofuels: production and applications

Unit 5: Microbiology of Soil, Water and Air (10 hrs)

- Concepts of habitat and niche, Microbial communities: nature, structure and attributes, levels of species diversity, Succession and stability, r and K selection.
- Ground water types and their contamination.
- Zonation of water ecosystem, Potability of water, Microbial assessment of water, Water purification.
- Air flora in different layers of atmosphere**,** Bioaerosol, Assessment of air quality using principles of Sedimentation, Impaction, Impingement, Suction and Filtration.
- Brief account of transmission of airborne microbes, Allergy: Causes and tests for detection of allergy.

Suggested Reading:

1. Agarwal, S. K. (1998). Environmental biotechnology. APH Publishing.
2. Alexander, M. (1999). Biodegradation and Bioremediation. Academic Press San Diego CA.
3. Chatterji, A. K. (2011). Introduction to Environmental Biotechnology. PHI Learning Pvt.Ltd..

4. Cookson Jr, J. T. (1995). Bioremediation engineering: design and application. McGraw-Hill, Inc..
5. Foster, C. F. (1987). John Ware DA, Environmental Biotechnology.
6. Jogdand, S. N. (2010). Environmental biotechnology.
7. Kamely, D., Chakrabarty, A., &Omenn, G. S. (1989). Biotechnology andbiodegradation. Gulf Publishing Co.

16PMBCC15	Practical (Core) –IV (Bioinformatics Practical)	3hrs/week	2Credits

1. Retrieval of biological sequences from major databases.
2. Editing of chromatogram, Elimination of contamination and submission of sequence to Genbank
3. Sequence Alignments – Pair wise & multiple sequence alignments
4. Sequence similarity search for a given sequence in biological databases using BLAST and FASTA.
5. Primer Designing
6. Characterization of a protein based on its primary structure using tools at EXPASY Molecular Biology Server
7. Retrieval of secondary/ tertiary structure of a protein from Protein Data Bank(PDB)
8. Protein Visualization (RASMOL, SPDB VIEWER, PROTEIN EXPLORER)
9. Predict Secondary structure of a protein using Chou & Fasman Method
10. Analysis of Ramachandran's plot for a protein structure.
11. Construction and evaluation of phylogentic tree for given sequences.