

**VANITA VISHRAM WOMEN'S UNIVERSITY**  
**SCHOOL OF SCIENCES**  
**DEPARTMENT OF MICROBIOLOGY**



**MASTER OF SCIENCE (M.Sc.) MICROBIOLOGY**  
**PROGRAMME**  
**under Learning Outcomes-based Curriculum Framework (LOCF)**  
**for Post Graduate (PG) Education**

**SEMESTERS 3**  
**Core Courses (CC)**

*Syllabus applicable to the students seeking admission in the*  
*M.Sc.- Microbiology*  
**under LOCF**  
**w.e.f. the Academic Year 2021-2022**



**SEMESTER 3  
CORE COURSE PAPER 8**

**MB21120 MOLECULAR BIOLOGY**

**Course Objectives:**

The purpose of this course is to introduce the student to the advanced concepts in molecular biology. Students will gain an understanding of molecular mechanisms of DNA replication, DNA repair, transcription, translation, and gene regulation in prokaryotic and eukaryotic organisms. The student will study the techniques and experiments used to understand these mechanisms.

**Course learning outcomes :**By the end of this course the students-

CO1: Is able to describe structure of DNA and RNA, organization of eukaryotic genome  
CO2: Is able to compare and contrast the mechanisms of bacterial and eukaryotic DNA replication, DNA repair, transcription

CO3: Is able to explain concepts in DNA repair mechanisms, and recombination as a molecular biology tool

CO4: Is able to explain various levels of gene regulation in both prokaryotic and eukaryotic organisms

CO5: Is able to describe post-transcriptional processes, RNA editing, RNAi and miRNA

CO6: Is able to describe translation mechanism in prokaryotes and eukaryotes, regulation of translation, and post-translational processing

CO7: Is able to describe post-translational processes

**THEORY COURSE  
(4+1 Credits)**

<b>Unit-1</b>	<b>The nature of Genetic material:</b> The structure of DNA and RNA; melting of DNA, superhelicity, organization of microbial genomes, organization of eukaryotic genomes, chromatin arrangement, nucleosome formation.	<b>15 Lectures</b>
<b>Uni-2</b>	<b>DNA replication:</b> Arrangement of replicons in a genome, various modes of replication, continuous, discontinuous synthesis, various replication enzymes, replication fork and priming, leading and lagging strand, elongation, termination, specific features of replication in prokaryotes and eukaryotes, action of topoisomerases, telomere maintenance and chromatin assembly, single stranded DNA replication, relationship between DNA replication and cell cycle, and DNA copy number maintenance.	<b>15 Lectures</b>

<b>Unit-3</b>	<b>Transcription &amp; Post-transcriptional processes:</b> Transcription machinery of prokaryotes, various transcription enzymes & cofactors, initiation, elongation & termination, sigma factors, transcription machinery of eukaryotes, various forms of RNA polymerase & cofactors, initiation, elongation and termination, promoters, enhancers, silencers, activators, effect of chromatin structure, regulation of transcription, RNA processing, splicing, capping and polyadenylation, rRNA and tRNA processing, RNA Editing; RNAi and miRNAs, Antisense RNA, Post transcriptional gene regulation.	<b>15 Lectures</b>
<b>Unit-4</b>	<b>Translation &amp; Post-translational processes:</b> The genetic code and protein structure, Mechanisms of translation in prokaryotes, Mechanisms of translation in eukaryotes, initiation complex, ribosomes and tRNA, factors, elongation and termination, <i>in vitro</i> translation systems, polycistronic/ monocistronic synthesis, Regulation of translation, RNA instability, inhibitors of translation, stringent response in bacteria, Protein modification, folding, chaperones, transportation. The Signal Hypothesis. Protein degradation.	<b>15 Lectures</b>
<p><b>Reference Book</b></p> <ol style="list-style-type: none"> <li>1. Molecular Biology by D.P. Clarke, N. Pazdernik. 2<sup>nd</sup> edition. Academic Press. 2012.</li> <li>2. Molecular Cloning: A laboratory manual by J. Sambrook, D. Russell. 4<sup>th</sup> edition. Cold Spring Harbor laboratory Press. 2012.</li> <li>3. DNA Technology: The Awesome Skill by I. Edward Alcamo. Harcourt Academic Press. 2001.</li> <li>4. Molecular Biology of the Gene by J. Watson, T. Baker, S. Bell, A. Gann, M. Levine, R. Losick. 7<sup>th</sup> edition. Pearson. 2014.</li> <li>5. Gene Cloning and DNA Analysis: An Introduction by T.A. Brown. 7<sup>th</sup> edition. Wiley Blackwell Publishers. 2016.</li> </ol>		

**SEMESTER 3  
CORE COURSE PAPER 9**

**MB21130 RECOMBIANAT DNA TECHNOLOGY**

**Course Objectives:**

The objective of this course is to make the student familiar with the currently used techniques to manipulate/ analyze DNA, RNA and proteins. The student will be made familiar with the methods used to clone genes, make and screen libraries, and the various applications of the polymerase chain reaction. The student will be taught about the methods currently used to carry out genome wide analyses and global analyses of transcription and protein expression. The student will be made familiar with how recombinant DNA technology has been exploited in the study of biology as well as in the production of pharmaceutical products.

**Course learning outcomes :**

Upon successful completion of the course, the student:

- CO1: Will be familiar with the use of various cloning vectors, and methods of DNA, RNA and protein analysis.
- CO2: Will be able to describe the various applications of PCR, and know how to make and screen genomic and cDNA libraries.
- CO3: Will be able to understand the methods by which DNA is sequenced and will gain insights into how entire genomes of organisms are sequenced.
- CO4: Will have learnt about promoter analyses, the many uses of the reporter genes, and methods to study the transcriptome.
- CO5: Will be aware of the different bacterial and eukaryotic systems available for overexpression of proteins.
- CO6: Will have learnt about different methods to analyze protein-DNA and protein-protein interactions, protein engineering, and methods for proteome analyses.
- CO7: Will know about the creation of plant and animal transgenics, and about animal cloning methods.

**THEORY COURSE  
(4+1 Credits)**

<b>Unit-1</b>	<b>Basics of DNA cloning, PCR and construction of cDNA and genomic DNA libraries:</b> Simple cloning and cloning using linkers and adaptors. Cloning into various kinds of vectors – plasmids, phages lambda and M13, phagemids, cosmids, P1 phage, PACs, BACs and YACs. Selection and screening of clones. Concept of PCR and various thermophilic enzymes used in PCR. Gradient PCR versus Touchdown PCR. Designing primers. Cloning PCR products. Long PCR, Inverse PCR, Vectorette PCR, RT-PCR, 5' and 3' RACE, Real Time PCR, Multiplex PCR. Steps in the construction of cDNA versus genomic DNA libraries. Screening libraries by colony hybridization and colony PCR.	<b>15 Lectures</b>
<b>Uni-2</b>	<b>Genome sequencing:</b> DNA sequencing by Sanger's method – traditional and cycle sequencing. Physical mapping by restriction fragment fingerprinting of BAC clones and STS mapping. E-PCR. Whole genome shotgun sequencing. Clone-by-clone shotgun sequencing of genome – preparation of BAC/YAC library, selection of BACs, subclone library construction, random shotgun phase and finishing phase followed by sequence authentication. Genome annotation at the nucleotide level, protein level and process level. Next	<b>15 Lectures</b>

	Generation sequencing methods.	
<b>Unit-3</b>	<b>Transcriptional analysis of gene expression and transcriptomics:</b> Gene expression analysis by Northern Blotting, RT-PCR, EST analysis and the use of reporter genes. Enzymatic and bioluminescent reporters. Reporters used in protein localization and trafficking studies. Promoter analysis – deletion analysis and linker scanning analysis coupled to reporter assays, mapping transcriptional start sites by S1 nuclease mapping, primer extension studies or 5' RACE. Transcriptome analysis by DD-PCR and EST analysis, DNA microarrays (cDNA arrays and oligo arrays), Serial Analysis of Gene Expression (SAGE), RNA-seq.	<b>15 Lectures</b>
<b>Unit-4</b>	<b>Analysis of protein-DNA and protein-protein interactions, protein engineering and proteome analysis:</b> Gel retardation assay, DNA footprinting by DNase I and chemical methods, yeast one-hybrid assay, ChIP- chip, ChIP-seq. Yeast two hybrid, three-hybrid, split hybrids and reverse hybrid. Co-immunoprecipitation, pull-down, far-western. Use of GFP and its variants in FRET analysis, use of BiFC. Phage display. Insertional and deletion mutagenesis. Site directed mutagenesis by conventional and PCR-based methods. Proteome analysis by 2D gel electrophoresis coupled to mass spectrometric analysis. Principles and use of MALDI-TOF and LC-MS platforms. PMF verses MS/MS. Protein arrays and their applications.	<b>15 Lectures</b>
<b>Reference Book</b> <ol style="list-style-type: none"> <li>1. Molecular Biology by D.P. Clarke, N. Pazdernik. 2<sup>nd</sup> edition. Academic Press. 2012.</li> <li>2. Molecular Cloning: A laboratory manual by J. Sambrook, D. Russell. 4<sup>th</sup> edition. Cold Spring Harbor laboratory Press. 2012.</li> <li>3. DNA Technology: The Awesome Skill by I. Edward Alcamo. Harcourt Academic Press. 2001.</li> <li>4. Molecular Biology of the Gene by J. Watson, T. Baker, S. Bell, A. Gann, M. Levine, R. Losick. 7<sup>th</sup> edition. Pearson. 2014.</li> <li>5. Gene Cloning and DNA Analysis: An Introduction by T.A. Brown. 7<sup>th</sup> edition. Wiley Blackwell Publishers. 2016.</li> </ol>		

### **MB21150: PRACTICAL V**

**Marks: 100 Duration: 60 hours (4 credits)**

**Course Objectives:**

The objective of this course is to train the student in basic molecular biology and microbial genetics

techniques. The student will learn how to isolate, analyze, and manipulate DNA, amplify DNA, fingerprint microbes, overexpress and purify recombinant proteins. The student will become familiar with transferring genetic material into bacteria by transformation and conjugation methods.

### **Course Learning Outcomes:**

The student:

CO1. Is able to perform restriction digestion and carry out its analysis by agarose gel electrophoresis.

CO2. Is able to perform restriction digestion and carry out its analysis by agarose gel electrophoresis.

CO3. Learns how to prepare competent cells and determine transformation efficiency

CO4. Learns how to do basic cloning

CO5. Is able to fingerprint microorganisms by RAPD analysis

CO6 Is able to overexpress recombinant proteins and analysis by SDS-PAGE

### **Contents:**

1. To determine molecular weight of a fragment from band profile.
2. To analyze plasmid DNA by restriction digestion followed by agarose gel electrophoresis.
3. To analyze plasmid DNA by restriction digestion followed by polyacrylamide gel electrophoresis.
4. To prepare competent cells by chemical method and determine their transformation efficiency
5. To isolate genomic DNA.
6. To amplify a gene of genomic DNA using PCR.
7. Blue white selection for identification of recombinant vector.
8. To perform RAPD analysis for microbial identification.
9. Cloning and Expression of a protein X
10. Separation of protein using SDS-PAGE analysis.
11. Determine molecular weight of a protein from gel pattern.

### **Suggested Readings:**

1. Molecular Cloning: A laboratory manual by Joseph Sambrook, David Russell, 4<sup>th</sup> edition. Cold

Spring Harbor laboratory Press. 2012.

2. Sequence - Evolution - Function: Computational Approaches in Comparative Genomics by E.V. Koonin , M.Y. Galperin. Kluwer Academic, USA. 2003.
3. Bioinformatics: A Practical Guide to the Analysis of Genes and Proteins edited by A. D. Baxevanis, B.F. Francis Ouellette . 3rd edition. Wiley and Sons. 2004.

## **MB21160: Practical VI**

### **Course learning outcome:**

The student:

- CO1. Can find ORFs in given nucleotide sequence using ORF Finder.
- CO2. Can create phylogenetic tree from the given nucleotide and protein sequence.
- CO3. Can perform protein modelling using SWISS-MODEL.
- CO4. Can analyse amino acid sequence to predict the structure and functions

### **Contents:**

1. Isolation of a hyper producing mutant by treatment of Physical/chemical mutagen.
2. To find ORFs in given nucleotide sequence using ORF Finder.
3. To create phylogenetic tree from the given nucleotide and protein sequence.
4. To Study Protein Parameters using Protparam.
5. To study domain architecture using ExPASy PROSITE.
6. To perform protein modeling using SWISS-MODEL.
7. To create multiple sequence alignments
8. To visualize and understand structures from PDB using PyMol/DeepView.
9. To predict secondary structure of a given amino acid sequence.
10. To analyzed protein properties from sequence- PROTPARAM

### **Suggested Readings:**

1. Molecular Cloning: A laboratory manual by Joseph Sambrook, David Russell, 4<sup>th</sup> edition. Cold Spring Harbor laboratory Press. 2012.
2. Sequence - Evolution - Function: Computational Approaches in Comparative Genomics by E.V. Koonin, M.Y. Galperin. Kluwer Academic, USA. 2003.
3. Bioinformatics: A Practical Guide to the Analysis of Genes and Proteins edited by A. D. Baxevanis, B.F. Francis Ouellette. 3rd edition. Wiley and Sons. 2004.



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**SEMESTERS 3**  
**Department Specific Elective (DE)**

*Syllabus applicable to the students seeking admission in the*  
*M.Sc.- Microbiology*  
**under LOCF**  
**w.e.f. the Academic Year 2023-2024**

## Structure of the Course

<b>Semester III</b>				
<b>Number of Department Elective Courses</b>	<b>Credits in each Elective Course</b>			
Course	Theory	Practical	Tutorial	Credits
MB24010: Biophysical and Biochemical Methods**	4	0	0	4
MB24030: Plant-Pathogen Interactions**	4	0	0	4
Elective course 'n'(total no) = 2	4	0	0	4
<b>Total credits in Elective Courses</b>	<b>4</b>			
<b>**Student must opt for any one of the Two Elective Courses</b>				

### Department Elective (DE)

1. MB24010: Biophysical and Biochemical Methods\*\*
2. MB24020: Advance Instrumental Microbiology
3. MB24030: Plant-Pathogen Interactions\*\*
4. MB24040: Food Microbiology\*\*
5. MB24050: Research Methodology
6. MB24060: Scientific Writing

# MASTER OF SCIENCE MICROBIOLOGY

## SEMESTER 3

### DEPARTMENT ELECTIVE COURSE PAPER 1

#### MB24010 BIOPHYSICAL AND BIOCHEMICAL METHODS

##### Course Objectives:

To introduce the student to the variety of biophysical and biochemical techniques currently available to probe the structure and function of the biological macromolecules, make them aware of the physical principles behind each technique and the instrumentation involved, make them familiar with various methods of analyzing the output data, and to build a strong foundation in the area of bacterial cell structure, division, survival and propagation.

##### Course learning outcomes :By the end of this course the students-

CO1: Be able to carry out the analysis of the data from CD and Fluorescence experiments to monitor the stability of the protein under different environmental conditions

CO2: Be familiar with the output of fluorescence and confocal microscopy

CO3: Be able to evaluate the quality and highlights of the structure reported/deposited in journals/structural databases.

CO4: Be able to design a multi-step purification protocol for a target protein

CO5: Be able to understand and correctly interpret the migration of protein molecule on PAGE under native and SDS conditions

CO6: Follow the safety precautions while using radioactive methods

#### THEORY COURSE (4 Credits)

<b>Unit-1</b>	<b>Microscopy:</b> Basics of microscopy: image formation, magnification, resolution, Biological applications and instrumentation of various kinds of microscopy: Optical Microscopy, Fluorescence, Confocal and Electron Microscopy, AFM, STM	<b>15 Lectures</b>
<b>Uni-2</b>	<b>Spectroscopy:</b> Various theories exploring the concept of light: Corpuscular theory, Wave theory, Electromagnetic theory, Planck's concept and modern theory. Basic concepts, principles and biological applications of different types of spectroscopy: absorption spectroscopy, fluorescence spectroscopy, phosphorescence, Infrared and Raman spectroscopy, Optical Rotatory Dispersion (ORD), Circular Dichroism (CD).	<b>15 Lectures</b>
<b>Unit-3</b>	<b>Separation Techniques: Chromatography:</b> Basics principles and applications of various chromatography methods: Partition and Absorption chromatography, gel filtration, ion exchange and affinity chromatography. Biological applications of HPLC and FPLC.	<b>15 Lectures</b>

<b>Unit-4</b>	<b>Radioactive methods:</b> Basics of radioactive isotopes and radioactive decay, sample preparation, counting, Safety precautions during handling, biological applications.	<b>15 Lectures</b>
<b>Reference Book</b>		
<ol style="list-style-type: none"> <li>1. Fundamentals of Molecular Spectroscopy by Colin Banwell. 4<sup>th</sup> edition. McGraw Hill.1994.</li> <li>2. Principles of Fluorescence Spectroscopy by J. Lakowicz, R. Joseph. 2<sup>nd</sup> edition.Springer.1999.</li> <li>3. Molecular Fluorescence: principles and Applications by B. Valeur. 2<sup>nd</sup> edition. Wiley. 2013.</li> <li>4. NMR – Conformation of Biological Molecules by G. Govil, R.V. Hosur. 1<sup>st</sup> edition. Springer- Verlag, 2011.</li> <li>5. Biomolecular crystallography: Principles, practice and application to structural biology by B. Rupp. 1<sup>st</sup> edition. Garland Science. 2009.</li> <li>6. Optical methods in Biology by E.M. Slayter. 1<sup>st</sup> edition. John Wiley. 1970. 7. NMR of proteins and nucleic Acids by K. Wuthrich. 1<sup>st</sup> edition. Wiley Interscience Publications. 1988.</li> <li>8. Biophysical chemistry, Part 2: Techniques by C. R. Cantor, P. R. Schimmel. 1<sup>st</sup> edition. W.H Freeman and Co. 2008.</li> </ol>		

**SEMESTER 3**  
**DEPARTMENT ELECTIVE COURSE PAPER 3**  
**MB24030 PLANT-PATHOGEN INTERACTIONS**

<p><b>Course Objectives:</b></p> <p>The course will facilitate in understanding of how pathogens interact with various plants and effect plant physiology, photosynthesis, respiration, transpiration and translocation. The involvement of various enzymes and toxins and understanding the molecular interaction will help in designing biocontrol strategies and development of transgenic plants. The course covers the novel molecular diagnostic approaches and correct forecasting of plant diseases.</p>
<p><b>Course learning outcomes :</b>By the end of this course the students-  Upon successful completion of the course, the student:</p> <p>CO1: Will have acquired knowledge about cause of plant diseases and effect of microbial infections on plant physiology, photosynthesis, respiration, transpiration, translocation</p> <p>CO2: Will have learnt about various enzymes and toxins in plant diseases and also role of phytoalexins.</p> <p>CO3: Understands about crown gall, symptoms of viral diseases and their control, diseases of some important cereals, vegetables and crops</p> <p>CO4: Will have gained insight into genetics of host-pathogen interactions, resistance genes,resistance mechanism in plants.</p>

<p>CO5: Will have been introduced to plant disease control, physical, chemical and biological methods of disease control</p> <p>CO6: Will have attained knowledge about designing of molecular diagnosis of plant disease and development of transgenic plants with applications and constraints.</p> <p>CO7: Is able to describe various important milestones in disease control and disease forecasting relevant in Indian farming.</p>		
<p><b>THEORY COURSE</b> <b>(4 Credits)</b></p>		
<b>Unit-1</b>	<p><b>Physiology and biochemical basis of plant diseases:</b> Causes of disease, pathogenesis, pathogenesis in relation to environment, effect of microbial infections on plant physiology, Enzymes and toxins in plant diseases, phytoalexins.</p>	<b>15 Lectures</b>
<b>Uni-2</b>	<p><b>Some important plant diseases and their etiological studies:</b> Crown gall, symptoms of viral diseases and their control, diseases of some important cereals, vegetables and crops.</p>	<b>15 Lectures</b>
<b>Unit-3</b>	<p><b>Disease control and forecasting:</b> Principles of plant disease control, physical and chemical methods of disease control, biocontrol, biocontrol agents - concepts and practices, fungal agents, <i>Trichoderma</i> as biocontrol agent, biocontrol agents – uses and practical constraints. History and important milestones in disease control, disease forecasting and its relevance in Indian farming.</p>	<b>15 Lectures</b>
<b>Unit-4</b>	<p><b>Genetic basis and Molecular approach of plant diseases:</b> Genetics of host-pathogen interactions, resistance genes, resistance mechanisms in plants, Molecular diagnosis, transgenic approach for plant protection, futuristic vision of molecular diagnosis, applications and constraints.</p>	<b>15 Lectures</b>
<p><b>Reference Book</b></p> <ol style="list-style-type: none"> <li>1. Plant Pathology by G. N. Agrios. 5<sup>th</sup> edition. Academic Press. 2005</li> <li>2. Plant Pathology by R.S. Mehrotra, and A. Aggarwal, 3<sup>rd</sup> edition. Tata McGraw Hill. 2017</li> <li>3. Bacterial plant pathology: cell and molecular aspects by D. C. Sige. Cambridge University Press.1993.</li> <li>4. Molecular plant pathology by M. Dickinson. BIOS Scientific Publishers, London. 2003.</li> <li>5. The essentials of Viruses, Vectors and Plant diseases by A.N. Basu&amp; B.K. Giri. Wiley Eastern Limited.1993.</li> <li>6. Biocontrol of Plant Diseases (Vol. I) by K.G. Mukerji and K.L.Garg. CRC Press Inc.,USA.1988.</li> <li>7. Molecular Biology of Filamentous Fungi by U. Stahl and P. Tudzyski. VCH VerlagsgesellschaftmbH. 1992.</li> </ol>		