

**Department of Microbiology**  
**(DST-FIST Supported, Centre of Excellence)**

The Masters of Science in Microbiology programme consists of 2 year (4 semesters).

The course details are as under:

<b>Paper</b>	<b>Course Title</b>	<b>Maximum marks</b>
<b>Semester I</b>		<b>Theory/Sessional</b>
<b>MB101</b>	General Microbiology	70/30
<b>MB102</b>	Bacterial Physiology	70/30
<b>MB103</b>	Structure, Function of Biomolecules & Analytical Methods	70/30
<b>MB104</b>	Microscopy, Biostatistics & Bioinformatics	70/30
<b>MBP105</b>	Practical I	200
	<b>Total Marks</b>	600
<b>Semester II</b>		
<b>MB201</b>	Agriculture & Environmental Microbiology	70/30
<b>MB202</b>	Biocatalysis, Bioenergetics & Metabolic pathways	70/30
<b>MB203</b>	Molecular Biology & Bacterial Genetics	70/30
<b>MB204</b>	Medical Microbiology	70/30
<b>MBP205</b>	Practical II	200
	<b>Total Marks</b>	600
<b>Semester III</b>		
<b>MB301</b>	Evolution & Microbial Taxonomy	70/30
<b>MB302</b>	Concepts of Immunology	70/30
<b>MB303</b>	Industrial and Food Microbiology	70/30
<b>MB304</b>	Modern Microbial Technology	70/30
<b>MBP305</b>	Practical III	200
	<b>Total Marks</b>	600
<b>Semester IV</b>		
<b>MB401</b>	Project work (Evaluation of Dissertation by external expert + Performance in the lab to be evaluated by the project supervisor)	500 (300+200)
<b>MB402</b>	Seminar presentation & viva-voce	100
	<b>Total Marks</b>	600
<b>Grand Total of Marks</b>		<b>2400</b>

## **Paper MB101 (Semester I)**

### **General Microbiology**

#### **Unit I**

History, development and scope of microbiology: Doctrine of spontaneous generation; Controversy over spontaneous generation; contribution of Antony van Leeuwenhoek, Lazzaro Spallanzani, Jhon Tyndall, Louis Pasteur, Joseph Lister, Iwanoski, Robert Koch in the development of microbiology. Microbiology in 20<sup>th</sup> Century.

#### **Unit II**

Nature of microbial world: General features of Eukaryotes and Prokaryotes. Differentiating features of Protists, Eubacteria and Archaeobacteria.

#### **Unit III**

Definition of nutritional groups of bacteria: Photoautotrophs, photoheterotrophs, chemoautotrophs, chemoheterotrophs. Microbial growth: Definition, measurement of growth, generation time, Arithmetic growth v/s exponential growth. Growth curve and growth phases. Diauxic growth. Synchronous and continuous cultures. Factors affecting growth: temperature; pH, oxygen.

#### **Unit IV**

Viruses: General properties and structure. Differences between viruses and cellular organisms. An introduction to mutations: Definition, physical and chemical mutations, physical mutagens (ionizing and non-ionizing radiations), chemical mutagens (base analogue, nitrous acid, hydroxyl amines, alkylating agents), Ames test.

#### **Unit V**

An introduction to microbial genetics: concepts of recombination in bacteria-transformation, conjugation and transduction.

**Paper MB102 (Semester I)**  
**Bacterial Physiology**

**Unit I**

Relation between structure and function in prokaryotic cells: cell wall, cell membrane, capsule, flagella, pili. Tactic movements; storage granules; metabolism of volutin (polyphosphates), glycogen, poly  $\beta$ -hydroxy alkanoates. Endospore structure and process of sporulation

**Unit II**

Physiology of extremophiles: thermophiles, psychrophiles, osmophiles (halophiles, saccharophiles), acidophiles and alkalophiles. Luminiscent bacteria and mechanism of luminiscence.

**Unit III**

Bacterial transport system: Donnan equilibrium, thermodynamics of transport systems, transport proteins, PEP-PTS system in relation to catabolite repression. Signal transduction and two component regulatory system.

**Unit IV**

Bacterial photosynthesis: Oxygenic and anoxygenic photosynthesis, photosystems, electron transport system, photophosphorylation, photorespiration.

**Unit V**

Structure of nitrogenase complex, mechanism of nitrogenase system, Symbiotic nitrogen fixation; ammonia regulation of nitrogenase; protection of nitrogenase against oxygen; nitrate and ammonia assimilatory enzymes.

**Paper MB103 (Semester I)**  
**Structure, Function of Biomolecules & Analytical Methods**

**Unit I**

Water: Structure, Physical and chemical properties. Handerson – Hasselbalch equation. Dissociation of water and its ionic product. Dielectric constant. pH and buffers. Van der waals , electrostatic, hydrogen bonding and hydrophobic interaction. Centrifugation: Introduction, principles and application of laboratory centrifuges with reference to differential, density gradient, preparative and analytical ultracentrifuges.

**Unit II**

Carbohydrates: Chemical structure, nomenclature, classification, properties and biological significance.

Amino acids and Proteins: Structure, properties, reaction of amino acids.

**Unit III**

Protein structure: Primary, secondary (Ramachandran plot), tertiary and quaternary structures. Protein designing, sequencing, targeting. Protein engineering.

Spectrophotometry: Ultra-violet, visible, AES/AAS, fluorimetry, polarimetry, circular dichroism, electron spin resonance, nuclear magnetic resonance, mass spectrometry, matrix assisted laser desorption/ionization (MALDI). pH metry. Biosensors.

**Unit IV**

Lipids: Function and properties of saturated and unsaturated fatty acids.

Biomembranes: Membrane structure and composition; membrane channels.

Nucleic acids: components of nucleic acids, structures and properties. Factors affecting structure of nucleic acid (melting curve, hydrophobic interactions, ionic strength).

Autoradiography- preparation, labeling, detection and measurement of radioactivity.

**Unit V**

Chromatography: Principles and application of gel filtration, high pressure liquid, ion exchange, affinity, gas- liquid, paper and thin layer chromatography.

Electrophoresis: Principles and application of polyacrylamide gel electrophoresis, 2-D gel electrophoresis, Isoelectric focusing and Agarose gel electrophoresis.

**Paper MB104 (Semester I)**  
**Microscopy, Biostatistics & Bioinformatics**

**Unit I**

**Microscopy:** Principles and application of light microscopy (bright field, dark field, phase contrast, and fluorescence microscopy, confocal Microscopy), electron microscopy (scanning and transmission electron microscopy).

**Unit II**

**Biostatistics:** Introduction to statistics: mean, median, mode, standard deviation, probability distribution, chi-square test, T-test, F-test.

**Unit III**

**Introduction to computers and bioinformatics:** Types of operating systems, concepts of networking and remote login, basic fundamentals of working with unix.

**Biological databases:** Overview, modes of database search, mode of data storage (Flat file format, db-tables), flatfile formats of GenBank, EMBL, DDBJ, PDB.

**Sequence alignment:** Concept of local and global sequence alignment, Pairwise sequence alignment, scoring an alignment, substitution matrices, multiple sequence alignment.

**Unit IV**

**Phylogenetic analysis:** Basic concepts of phylogenetic analysis, rooted/uprooted trees, approaches for phylogenetic tree construction (UPGMA, Neighbour joining, Maximum parsimony, Maximum likelihood).

**Generation and analysis of high throughput sequence data:** Assembly pipeline for clustering of HTGS data, format of “.ace” file, quality assessment of genomic assemblies, International norms for sequence data quality, Clustering of EST sequences, concept of Unigene.

**Unit V**

**Annotation procedures for high through-put sequence data:** Identification of various genomic elements (protein coding genes, repeat elements, strategies for annotation of whole genome, functional annotation of EST clusters, gene ontology (GO) consortium.

**Structure predictions for nucleic acids and proteins:** Approaches for the prediction of RNA secondary and tertiary predictions, energy minimization and base covariance models, Basic approaches for protein structure predictions, comparative modeling, fold recognition/ “threading” and *ab-initio* prediction.

**Practical I**  
**MBP105 (Semester I)**

- Acquaintance with microbiology laboratory and equipments.
- Techniques in Microbiology: Sterilization methods, Isolation of pure culture, Cultivation of aerobic and anaerobic bacteria, Preservation and maintenance of microbial cultures.
- Calibration of micrometer and bacterial size measurements.
- Structure and morphological features of yeast, molds, bacteria, algae, protozoa, and actinomycetes.
- Staining techniques in bacteria- simple, negative, Gram and special-endospore and capsule.
- Staining of nuclear material of Bacteria.
- Hanging drop technique for demonstrating bacterial motility.
- Fungal isolation and identification.
- Identification of the product of metabolic pathways in isolated bacterial culture-carbohydrate catabolism, catalase and peroxidase activities, IMV ic test, gelatin and starch hydolysis, cellulose degradation and H<sub>2</sub>S production.
- Estimation of thermal death point (TDP) and thermal death time (TDT) of microorganism.
- Estimation of nitrate reductase.
- Study of diauxic growth curve.
- Microbial examination of reserve materials.
- Determination of pK<sub>a</sub> of amino acids.
- Qualitative determination of sugars by Benedict's and Fehling's test.
- Estimation of glucose by Anthrone/ Nelson Somogyi method.
- Estimation of polysaccharides –starch, cellulose and glycogen.
- Estimation of protein by Lowery and Biuret methods.
- Estimation of nucleic acids.
- Measurement of saponification value of lipids.
- Chromatography: paper Chromatography (ascending, descending).
- Thin layer Chromatography.
- Column Chromatography (Gel Exclusion, Ion Exchange, Affinity).
- Demonstration of PAGE, SDS-PAGE, agarose gel electrophoresis.

**Paper MB201 (Semester II)**  
**Agriculture & Environmental Microbiology**

**Unit I**

Soil environment: Distribution, abundance, Common methods of enumeration and maintenance of soil microflora. Effect of pesticides on microbial population. Transformation of carbon, nitrogen and phosphorus. Mineralization and immobilization of nitrogen: ammonification, nitrification, denitrification, nitrogen fixation (symbiotic and nonsymbiotic) and its significance in agriculture.

**Unit II**

Biofertilizers: their methods of preparation and seed bacterization. Humus formation. Microbiology of composting. Microbial associations: Symbiosis, Synergism, Commensalism, Antagonism, Parasitism, Competition. Host-microbe interaction: rhizosphere, phyllosphere, mycorrhiza, Plant growth promoting rhizobacteria (PGPR), Siderophores in relation to rhizobacteria.

**Unit III**

Microbiology of air including sampling techniques, role in air pollution and sanitation. Microbiology of water: Distribution and techniques. Algal bloom. Microbiology of water / waste water and treatment systems. Biogas generation.

**Unit IV**

Microbial decomposition of organic matter – cellulose, hemicellulose and lignin. Degradation of pesticides, Xenobiotics and plastics. Biodegradable plastic and biopesticides.

**Unit V**

Microbial adaptation to environmental stress. Biomagnification and bioremediation. Genetically engineered microbes in environment. Effect of radiation and Green house effect.

**Paper MB202 (Semester II)**  
**Biocatalysis, Bioenergetics and Metabolic Pathways**

**Unit I**

Enzymes: Introduction, basic properties and classification. Kinetics: negative and positive cooperativity, Michalis-Menton equation. Enzyme inhibition, active sites, allosteric enzymes, isozymes, ribozymes, deoxy ribozymes, abzymes and artificial enzymes.

Structure and biochemical functions of vitamins, coenzymes and hormones.

**Unit II**

Biochemical energetics: laws of thermodynamics; concepts of energy. Oxidation-reduction potential. Hydrolysis of energy rich intermediates. Photophosphorylation and pigment systems.

Respiratory electron transport: components and inhibitors of the electron transport chain, energy transduction and proton-motive force, chemiosmotic theory of ATP generation.

**Unit III**

Metabolism and regulation of carbohydrates: Glycolysis, krebs cycle, pentose phosphate pathway, gluconeogenesis, glycogenesis, starch synthesis. Hormonal regulation of carbohydrate metabolism.

**Unit IV**

Amino acid biosynthesis and degradation. Urea cycle.

Biosynthesis of proteins and their regulation. Protein engineering.

**Unit V**

Fatty acid metabolism:  $\alpha$ ,  $\beta$ -oxidation and biosynthesis of fatty acids.

Purine and pyrimidine biosynthesis by de novo pathway. Catabolism of nucleotides.

**Paper MB203 (Semester II)**  
**Molecular Biology & Bacterial Genetics**

**Unit I**

Concepts of nucleic acids as genetic material, genome organization (chromatin structure, coding and noncoding sequences, satellite DNA). C-value paradox, Cot analysis. Repetitive DNA, circular and superhelical DNA.

**Unit II**

DNA replication: general principles, types and properties of DNA polymerase, DNA replication mechanism. Gene transcription: types and role of polymerases in transcription, mechanism of transcription.

**Unit III**

Concept of reverse transcriptase. Protein synthesis and processing: concepts and properties of genetic code, central dogma, tRNA processing, translation of mRNA into protein. Cistron, recon, muton. Genomics, transcriptomics and proteomics.

**Unit IV**

Mutation: Types of mutation, molecular basis of mutagenesis, Mutant reversion. DNA damage and repair. Regulation of gene expression: positive and negative control, induction, repression. Operon concepts- *lac*, *ara*, and *trp* operons.

**Unit V**

Properties and replication of plasmids, Plasmid incompatibility. Bacterial recombination: transformation, conjugation and transduction. Restriction and modification. Gene mapping. Global aspects of catabolite repression and other global network.

## **Paper MB204 (Semester II)**

### **Medical Microbiology**

#### **Unit I**

History of medical microbiology. Normal microflora of human body. Medically important microorganisms with respect to history, pathogenicity, epidemiology, prophylaxis, treatment- *Staphylococcus*, *Streptococcus*, *Pneumococcus*, *Nisseria*, *Corynebacterium*, *Clostridium*, *Enterobacteriaceae*, *Pseudomonas*, *Yersinia*, *Haemophilus*, *Mycobacterium*, *Spirochaete*, *Rickettsia* and *Chlamydiae*.

#### **Unit II**

General characteristics and diseases caused by adeno-, pox-, influenza-, herpes-, rhabdo-, hepatitis-, polio-, rabies-, HIV-, MMR and oncogenic-viruses.

#### **Unit III**

Pathogenic fungal diseases: (i) Dermatophytes- *Trichophyton*, *Microsporum*, *Epidermophyton*. (ii) Filamentous fungi causing subcutaneous infection-*Phialophora*, *Madurella*, *Sporotrichum*, *Mucor*, *Rhizopus*, *Aspergillus*. (iii) Systemic mycoses- *Histoplasma*, *Blastomyces*. (iv) Yeast-like fungi- *Cryptococci*, *Candida*.

#### **Unit IV**

Diseases caused by protozoa: *Plasmodium*, *Leshmania*, Entamoebiasis. Use of animals as experimental models.

#### **Unit V**

Principles of chemotherapy: role of antimicrobial agents-synthetic compounds and antibiotics, their mechanism of action, drug resistance in bacteria, drug sensitivity test.

## **Practical II**

### **MBP205 (Semester II)**

- Isolation and enumeration of soil and air microflora.
- Isolation of free-living nitrogen-fixing bacteria.
- Isolation of rhizobia from root nodules and *Azospirillum* from grass.
- Isolation of actinomycetes and thermophilic microbes from soil.
- Demonstration of mycorrhiza in plant roots.
- Microbial analysis of water by most probable number (MPN) technique and presumptive, confirmatory and completed tests for coliforms.
- Estimation of BOD/COD in wastewater /effluent, before and after treatment.
- Demonstration of liberation of  $\text{NH}_3$  from nitrogenous organic compound.
- Effect of temperature and pH on bacterial growth.
- Study of enzyme kinetics.
- Lethal action of U.V. light and photoreactivation .
- Isolation and characterization of bacterial plasmid, DNA and plasmid curing studies.
- Determination of MIC for different antibiotics.
- Determination of heavy metal tolerance in Bacteria.
- Genetic transformation of antibiotic resistance.
- Collection and processing of samples for isolation and identification of pathogenic microorganisms.
- Isolation of pathogens from stool, pus, urine, mouth, nasal passage and their antibiotic sensitivity test.
- Identification of enteric pathogen by triple sugar iron agar (TSIA) test.
- Examination of teeth crevices for microbial flora.
- Determination of dental caries susceptibility.
- Separation serum; WBC and RBC count.
- Demonstration of Acid fast and acid alcohol fast staining for tuberculosis bacillus.

**Paper MB301 (Semester III)**  
**Evolution & Microbial Taxonomy**

**Unit I**

Classification of Bacteria, Actinomycetes, Cyanobacteria, Mollicutes, Rickettsia, Chlamydiae, Fungi and Slime molds. Classification and Phylogeny of Bacteria: Polyphasic approaches of bacterial Taxonomy, Numerical taxonomy, 16 s RNA analysis, etc. Significance and potential applications of various groups of bacteria according to Bergey's Manual of Systematic Bacteriology.

**Unit II**

Implications of molecular and biochemical methods including rDNA analysis, RFLP, RAPD and other fingerprinting techniques, fatty acids, polysaccharides and lipids and role of secondary metabolites in systematics.

**Unit III**

Emergence of Evolutionary thoughts. Lamarck Darwin- concepts of variation, adaptation, natural selection, Mendelism

**Unit IV**

Origin of cells and unicellular evolution. Origin of basic biomolecules, Abiotic synthesis of organic monomers; Concepts of Oparin and Haldane; Experiments of Miller (1953); The first Cell, Evolution of prokaryotic and eukaryotic cell.

**Unit V**

Molecular evolution & their mechanisms. Population genetics, Concepts and rate of change in gene frequency through natural selection, random genetic drift, Adaptive radiation, Speciation, Convergent evolution.

**Paper MB302 (Semester III)**  
**Concepts of Immunology**

**Unit I**

History of immunology. Cells and organs of immune system. Active & passive immunity. Antigens, antigenicity & immunogenicity. Adjuvant & haptens. Toxins & Toxoids.

**Unit II**

Structure and function of antibody molecules, generation of antibody diversity, antibody engineering. Hybridoma and monoclonal antibodies.

**Unit III**

Non-specific immunity: role of normal microflora of body, physical & physiological barriers, phagocytosis, inflammation, complement system, interferon. Specific immunity: humoral & cell mediated immunity.

**Unit IV**

Major histocompatibility complex and graft rejection. Antigen processing and presentation. Development of 'b' & 't' cells.

**Unit V**

Comparative study of Type I-V hypersensitivities with examples. Immunodeficiency diseases- defects of T cells, B cells, complement and phagocytic cells, etc. Immunotherapy & vaccines.

**Paper MB303 (Semester III)**  
**Industrial and Food Microbiology**

**Unit I**

Bioprocess technology: Quality of substrates and microorganisms for industrial applications, fermentor systems, scaling up, fermentation process, Types of fermentation, online monitoring and down-stream processing, Solid state and submerged fermentation, Use of computers in the fermentation process.

**Unit II**

Microbial production of ethanol; organic acids- citric and lactic acid; amino acids - glutamic acid, lysine; enzymes- amylases, proteases and lipases; vitamins- riboflavin; antibiotics (natural and synthetic)- penicillin, methicillin, streptomycin, bacitracin, tetracyclin; hormones; vinegar, wine, beer and polysaccharides.

**Unit III**

Microbial deterioration of industrial products (leather, cotton, paint, wood, etc.) and their control. Fermentation of tea, coffee, cocoa and tobacco.

Microbes as food and feed: mushroom, single cell protein (substrate, organisms, advantages, disadvantages and economic aspects of SCP), silage microbiology.

**Unit IV**

Microbial transformation of antibiotics and steroids. Microbial leaching of metal ores and desulfurization of coal. Immobilization of microbial cells/enzymes and their uses in various processes.

**Unit V**

Introduction to food microbiology, common food spoiling, intoxicating and disease causing microorganisms. Factors affecting microbial growth in foods. Antimicrobial constituents of foods. Microbial indicators of food safety. Physical, chemical and biological principles of preservation of foods (cereals, milk, meat, vegetables and fruits etc.). Canned food. Lactose peroxidase system, lactic antagonism. Food fermentation (milk, meat, fruit and vegetables). Therapeutic use of fermented food. Food quality systems and their quality control regulations.

## **Paper MB304 (Semester III)**

### **Modern Microbial Technology**

#### **Unit I**

Introduction to modern microbial technology and its various implications (in human therapeutics, agriculture, waste water treatment, etc.). Restriction endonucleases, plasmid vectors (natural and artificial), binary vectors, shuttle vector, vectors (their design and rationale). Bacteriophage  $\lambda$  vectors, cosmid & phagmid vectors and yeast vectors. DNA ligase and ligation.

#### **Unit II**

*In vitro* synthesis of DNA, genome sequencing, PCR: types and their uses. Thermostable enzymes- Taq and Pfu. Blotting techniques- Northern, Southern and Western blotting. Dot and slot blot techniques. Restriction fragment length polymorphism (RFLP) and its application.

Cloning and screening strategies, cloning in gram-positive, gram-negative bacteria and in yeast.

#### **Unit III**

**Synthesis of commercial products using microbial systems-** proteins: insulin, interferon and growth hormone, restriction endonucleases. Strategies of novel antibiotics (e.g. actinorhodine, medermycin, granaticin)- synthesis in streptomyces strains. Biopolymers- xanthane gum and PHAs. L-ascorbic acid (vitamin C). Microbial enzymes- lipase, acid and alkaline proteases.

#### **Unit IV**

**Production of vaccines and therapeutic agents-** subunit vaccines against *Herpes simplex* and foot & mouth disease, peptide vaccines, live recombinant vaccines, attenuated vaccines (cholera and salmonella). Vector vaccines (against viruses and bacteria). Monoclonal antibodies as therapeutic agents. Therapeutic agents against HIV.

#### **Unit V**

**Bioremediation-** degradation of xenobiotics and genetic engineering of biodegradative pathways.

**PGPR-** genetic engineering of nodulation genes, nitrogenase, and hydrogenase, Siderophores, gene clusters.

**Microbial insecticides-** *Bacillus thuringiensis* and its genetic engineering.

Use of *Agrobacterium tumefaciens* in transgenic plants.

**Practical III**  
**MBP305 (Semester III)**

- Isolation and characterization of bacterial chromosomal DNA and RNA
- Use of PCR to clone specified gene from bacteria.
- PAGE/SDS-PAGE of bacterial proteins.
- Serological test: blood group, Widal, VDRL, pregnancy test.
- Determination of ELISA, Immuno-diffusion, Immuno-electrophoresis techniques.
- Isolation of cellulose degrading microbes.
- Field visit to various industries for collection of sample for further analysis, isolation of cultures and purification of dominant microbes.
- Isolation and identification of microorganism from spoiled food, milk/milk products.
- Direct microscopic count of bacteria in milk and examination of pasteurized and sterilized milk.
- Preparation of Yoghurt and Sour-krat.
- Platform test of milk-MBR test, resazurine reduction test.
- Isolation and enumeration of microorganisms from decaying materials of commercial value.
- Production of alcohol from molasses and determination of fermentation efficiency.
- Production and estimation of industrially important enzymes-amylase, cellulase, protease.