Khandesh College Education Society's

Moolji Jaitha College, Jalgaon

An "Autonomous College" Affiliated to

KBC North Maharashtra University, Jalgaon

SYLLABUS

Microbiology

M. Sc. II (Semester III & IV)

Under Choice Based Credit System (CBCS)

[w. e. f. Academic Year: 2020-21]

M. Sc. Part: II Microbiology

Program Specific Outcomes (PSO)

Students who graduate with a Master of Science in Microbiology will:

- **PSO1**: Acquired knowledge and understanding of the microbiology concepts as applicable to diverse areas such as industrial, agriculture, food, environment, and others.
- **PSO2:** Demonstrate an understanding of structure and metabolism of macromolecules, regulation of metabolic pathways and the role of microbes in industry, health and environment.
- **PSO3:** Demonstrate key practical skills/competencies in working with microbes for study and use in the laboratory as well as outside, including the use of good microbiological practices.
- **PSO4:** Competent enough to use microbiology knowledge and skills to analyse problems involving microbes, articulate these with peers/ team members/ other stake holders, and undertake remedial measures/ studies etc.
- **PSO5:** Developed a broader perspective of the discipline of Microbiology to enable him to identify challenging societal problems through research and plan his professional career as entrepreneur to develop innovative solutions for such problems.

Course Structure: M.Sc. (Microbiology) (CBCS) Semester III and IV

Duration: The duration of M.Sc. (Microbiology) degree program shall be two years divided into four semesters. Each semester consists of 90 working days.

Semester	Course Module	Subject Code	Title of Paper	Credit	Hours per Week
III	DSC	MB-301	Applied and environmental microbiology	4	4
	DSC	MB-302	Pharmaceutical microbiology	4	4
	DSC	MB-303	Practical course in biotechnology	4	8
	DSC	MB-304	Practical course in applied microbiology	4	8
	SEC	MB-305	Research methodology	4	4
	DSE	MB-306	Agricultural microbiology	4	4
IV	DSC	MB-401	Fermentation technology	4	4
	DSC	MB-402	Applied molecular biology	4	4
	DSC	MB-403	Practical course in biostatistics and bioinformatics	4	8
	DSC	MB-404	Project dissertation	4	8
	GE	MB-405	Biostatistics and bioinformatics	4	4
	DSE	MB-406	Entrepreneurship in microbiology	4	4

DSC: Discipline Specific Elective Core Course; **SEC**: Skill Enhancement Course;

DSE: Discipline Specific Elective (DSE) Course; **GE**: Generic Elective Course

MB-YSC : Microbiology (Y-year; S-Semester; C-Course number)

Examination Pattern for M.Sc.

Examination	Marks
External Marks	**60**
Internal Marks	**40**
Total Marks	**100**

M.Sc. (Microbiology): Semester-III

MB-301: Applied and environmental microbiology

Hours: 60 **Credits: 4**

Course objectives:

- To understand strategies used for microbial analysis of food.
- To learn principle and methods of wastewater treatment.
- To impart knowledge about recalcitrant from contaminated environment and bioremediation

Course outcomes: On completion of this course, the student will be able to:

- Aquent with skills related microbial analysis of food and waste water treatment
- Understand use of microbes in solid and liquid waste treatment as well as bioremediation
- Explore the strategies of microbial waste management.

Unit I: Food Microbiology **(12 h)**

- Methods of sampling : random, representative, attribute sampling
- Preparation of dilutions
- Offline and online approaches of microbial analysis
- Detection and enumeration of indicator bacteria, pathogenic and toxigenic microbes
- Mycotoxins, sources, pathogenesis, prevention, extraction and detection
- Microbiological examination of specific foods
    - Meat and meat products, Milk and milk products
- Food intoxications: Causes, pathogenesis, prevention and control

Unit II: Microbiological treatment of waste water **(12 h)**

- Need for biological waste water treatment
- Overview of conventional treatment process: coagulation, flocculation, sedimentation, filtration, disinfection
- Primary: screen, grit chamber, primary sedimentation or settling
- Secondary biological treatment process:
    - Aerobic treatment: (a) Suspended growth - Oxidation lagoons, activated sludge and membrane bioreactor (b) Attached growth TF, RBC, PBR Anaerobic treatment: (a) Suspended growth - UASB, USB, Clarigester (b) Attached growth (EGSB, AF, FBR)
    - Disinfection (Clarification, Ozonation)
- Advanced tertiary process: biological removal of nitrogen and phosphorus,
- Permissible limit for domestic and industrial waste
- Waste water treatment for breweries and antibiotic industries

Unit III: Biological conversion of Lignocellulosic waste **(12 h)**

- Structure and composition of lignocelluloses,
- Degradation of lignin, cellulose and hemicellulose, impediments of degradation
- Pre-treatment of lignocellulosic material: Physical, Chemical and Biological (Microbial and enzymatic) and related issues

- Fermentation of lignocellulosic biomass: Submerged, SSF, SHF, SScF
- Applications of lignocellulosic waste for ethanol production

Unit IV: Bioremediation and biodegradation of xenobiotics (12 h)

- Concept of biodegradability and bioconversion
- Principles for measuring biodegradability
- Factors affecting biodegradability
- Mechanism of biodegradation / bioremediation: co-metabolism, mineralization, conjugation
- Strategies of Bioremediation : Ex-situ and in-situ
- Bioremediation treatments: intrinsic, biostimulation, and bioaugmentation, rhizostimulation, bioleaching, phytoremediation
- Methods for microbial treatments of pollution: bioreactors, biopiles, landfilling, bioventing, biosparging
- Limitations to microbial degradation of compounds
- Biodegradation of xenobiotics
    - Types and hazards of xenobiotics
    - Biochemical/ physiological approach
    - Molecular techniques and monitoring of bioremediation

Unit V: Microbial waste management (12 h)

Solid waste management

- Composting:Principle, steps, chemistry, microbial succession andbiology of composting, technologies of composting (aerobic: windrow, aerated static pile, in vessel composting; Anaerobic composting) criteria of compost maturity, applications of compost
- Anaerobic digestion: Feedstocks, Principle, methane potential,requirements of anaerobic digestion, microbiology of biomethanation, biochemistry of methane synthesis, process, dry and wet digestion, factor affecting methanogenesis, and types of anaerobic digesters (fixed dome, floating dome, low rate and high rate digesters)

References:

- Singh A and Ward O.P. (2004) Biodegradation and Bioremediation, Springer-Veil ag,, Berlin (ISBN: 3-540-21101-2).
- Hurst C.J. (2002) Manual of Environmental Microbiology, ASM Press, Washington D.C. (ISBN: 1-55581-199-x).
- Demain A.L. and Davies J.E. (1999) Manual of Industrial Microbiology and Biotechnology, ASM Press, Washington D.C. (ISBN: 1-55581-128-0).
- Martin A.M. (1998) Bioconversion of waste materials to Industrial Products, Blackie Academic and Professional, London (ISBN: 0-7514-0423-3).
- Harrigan W.F. and McCance M.E. (1994) Laboratory Methods in Food and Dairy Microbiology. Academic Press, London.
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- Hobbs B and Roberts D (1993) Food Poisoning & Food Hygiene, Edward Arnold,

London.
- Baker K.H. and Herson D.S. (1994) Bioremediation, Mc-Graw Hill Inc., New York.
- Pandey A. (2004) Concise Encyclopedia of Bioresource Technology, Food Products Press, The Haworth Reference Press, New York (ISBN: 1 -56022-980-2).
- Rehm R.G. and Reed G (1984) Biotechnology, Vol.1-8, Verlag-Chemie, Weinheim.
- Forster C.F. (1985) Biotechnology and waste water treatment, Cambridge University Press, Cambridge.
- Maier R Pepper I.L. and Gerba C.P. (2000) Environmental Microbiology, Academic Press, London.
- Rajendran P and Gunasekaran P (2006) Microbial Bioremediation, MJP publishers Chennai (ISBN: 81-8094-022-5).

M.Sc. (Microbiology): Semester-III
MB-302: Pharmaceutical microbiology

Hours: 60 **Credits: 4**

Course objectives:
- To introduce knowledge about antimicrobial agents and biopharmaceuticals.
- To impart the information related to regulatory aspects such as GMP, GLP, ICH process
- To familiarize the students with aspects of drug aspects of design

Course outcomes: On completion of this course, the student will be able -
- Get in-depth knowledge on mechanisms of different antimicrobial agents and biopharmaceuticals
- Understand quality control and regulatory aspects used in pharmaceuticals
- Discriminate conventional and combinatorial tools used in drug design and discovery.

Unit I: Antibiotics and Synthetic antimicrobial agents (12 h)
- Mechanism of action, microbial resistance, therapeutic, prophylactic usage and adverse reactions
- Antibiotic and Synthetic antimicrobial agents: beta-lactam, aminoglycosides, tetracyclines, ansamycins, macrolides
- Antifungal antibiotics: Griseofulvin
- Antiviral drugs: Amantidines, Nucleoside analogues, Interferons
- Peptide antibiotics
- Synthetic antibiotics: Sulphonamides, Chloramphenicol,
- Quinolone
- Anticancer drugs

Unit II: Regulatory aspects and quality assurance in pharmaceuticals (12 h)
- Regulations for pharmaceutical industry and pharmacopeia
- Lab design: General design, sample collection and testing, equipment, utility and service, air supply, contaminants, laboratory safety
- Laboratory management : Training, quality, safety, LIMS
- GMP in pharmaceuticals

    - US Food and Drug Administration (FDA) and European Union (EU GMP)
    - Key aspects of GMP compliance and rules of GMP
    - Risk management and GMP documentation
- Design of sterile product manufacturing unit: Class A and clean room
- Quality control in pharmaceuticals:
  In-process and final product control (antibiotics, immune sera, vaccines, biopharmaceuticals)
- ICH process
- Sterilization control and sterility validation
- In vitro and in vivo testing for pyrogens and endotoxins

Unit III: Microbiology laboratory techniques (12 h)

- Good laboratory practice (GLP), laboratory safety and aseptic technique
- Pharmacopeia and microbiological tests
- Microbiological examination of nonsterile products:
    - TVC: Bioburden determination, method validation, media growth promotion, sample preparation, test method (membrane filtration, pour / spread plate, MPN)
    - Tests for specified organisms and specification limits
    - Measurement of cell concentration in suspension by optical density
    - Sterility testing
    - Environmental monitoring and Water analysis

Unit IV: Production of Biopharmaceuticals (12 h)

- Asparaginase, and Clinical dextran
- Vaccines (DNA/ multivalent subunit/ bacterial)
- Viral vaccines: Live attenuated, Inactivated, , Live recombinant,Virion subunit vaccines, production of viruses for vaccines, Virus like particles, Synthetic peptide vaccines,
- Immunosera and monoclonal antibodies

Unit V: Drug design (12 h)

- Rational drug design
    - Concept and steps of drug design
    - Lead drug and Pro-drug with examples
    - Structure based and combinatorial approach
    - Computer aided drug design and softwares
    - Peptidomimetic
    - Strategies for drug discovery and clinical trial
- Drug delivery: Concept and methods of drug delivery, novel methods

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- Hugo W.B. and Russell A.D. (2003/1998) Pharmaceutical Microbiology, 6th edn, Blackwel Science, Oxford, UK (ISBN: 0-632-04196-X) Reprinted.

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M.Sc. (Microbiology): Semester-III

MB-304: Methods in Biotechnology (Practical Course)

Hours: 120 **Credits: 8**

Course objectives:

- To train the student in basic molecular biology tools
- To learn techniques in biogasanalysis and estimation of antibiotics
- To introduce microbial interaction with plant and rhizoshere

Course outcomes: On completion of this course, the student will be able to:

- Perform the basic molecular techniques related to RNA and DNA
- Understand plant microbe relations with plant and rhizoshpere
- Use the methods in biogas analysis and estimation of antibiotics

List of practicals

1. Isolation and estimation of RNA / mRNA from bacteria/ yeast/ fungi
2. Determination of Tm and % (G+C) of DNA
3. DNA fingerprinting through southern blotting
4. Biotransformation of antibiotic/ steroid
5. Determination of efficacy of any suitable antibioticby microbiological and chemical assay
6. Preparation of SOP for laboratory instrument /procedure (any one Autoclave/Spectrophotometer/ oven / incubatory/balance/ etc)
7. Fermentative production of alcohol from lignocellulose and its estimation
8. Analysis of biogas digested slurry for organic C, N, COD, lignin, Fatty acids
9. Nodulation of legume by Rhizobium using Leonard Jar/ Pot assay
10. Production and detection of siderophore produced by bacteria / fungi
11. Isolation of VAM spores from soil
12. Isolation of microbes from Rhizosphere / Phyllo-plane/ PGPR
13. Estimation of R:S ratio and assessment of rhizosphere effect.

References:

- Aneja KR (2007). Experiments in microbiology, plant pathology and biotechnology. New Age International, New Delhi
- Benson HJ (2002). Microbiological applications; a laboratory manual in general microbiology, 8th Edition, McGraw Hill, New York
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- White D (2000) The Physiology and Biochemistry of Prokaryotes, Oxford University Press, Oxford.

M.Sc. (Microbiology): Semester-III

MB- 304: Methods in applied microbiology (Practical Course)

Hours: 120 **Credits: 8**

Course objectives:

- To impart training about aspects of microbiological quality control at pharmaceutical industry
- To familiarize with quality activities required in pharmaceutical industry
- To understand the methods of saccharification of lignocelluosic biomass

Course outcomes: On completion of this course, the student will be able to:

- Undertake various quality control tests of pharmaceutical products and media
- Carry out sterility testing of pharma products and perform the validation of LAF
- Understand various methods used for saccharification of lignocellulosic biomass

1. Validation of autoclave using chemical and biological indicator
2. Validation of efficiency of laminar air flow
3. Survivor curve for Ultraviolet light/Heat /ethylene oxide and D, Z. F value determination

4. Environmental monitoring of samples from production areas and personnel.
5. Evaluation of disinfectant using Phenol coefficient (Rideal Walker Test/ Chick Martin Test)
6. Sterility testing of in-process materials and finished products
7. Microbial Limit Test: analysis of water/ raw material/ finished product/ packaging material/ Excipients)
8. Evaluation of quality of media/reagents for growth promotion tests.
9. Evaluation of carcinogenicity using Ames test
10. Endotoxin/ pyrogen using LAL (water/ in-process/ final product)
11. Evaluation of microbial spoilage of refrigerated and canned food
12. Lignocellulosic saccharification using SSF/SHF/SScF and its assessment
13. Production of biogas using feedstock and detection using water displacement method/ GC

References:

1. White D (2000) The Physiology and Biochemistry of Prokaryotes. Oxford University Press. Oxford.
2. Mudili J (2007) Introductory Practical Microbiology, Narosa Publ. House Pvt. Ltd. New Delhi (ISBN: 978-81-7319-744-4).
3. Primrose SB and Wardlow AC (1982) Source Book for Experiments for the Teaching of Microbiology, Academic Press, London (ISBN: 0-12-565680-7).
4. SawhneySk and Singh R (2001) Introductory Practical Biochemistry, Narosa Publ. House. Chennai.

M.Sc. (Microbiology): Semester-III

MB 305: Research methodology

Hours: 60 **Credits: 4**

Course objectives:

- To acquaint the student with fundamental research
- To inculcate techniques of research process
- To introduce with technique of research documentation and anti-plagiarism

Course Outcomes: On completion of this course, the student will be able to:

- Perform the research with systematic and scientific approach
- Understand research process, formulate research plan and analyse the data
- Use the methods of report writing and checking plagiarism

Unit I: Fundamentals of research **(12 h)**

- Objectives of research
- Type of research:
    - Descriptive vs. Analytical, Applied vs. Fundamental, Quantitative vs. Qualitative, Conceptual vs. Empirical, Experimental Research Exploring or Formulative Research, Diagnostic Research/Study, Surveys, Case Study, Field Studies
- Criteria for good research
- General research process

- Define the research problem, literature survey, formulating hypotheses, research design, data collection and analysis, interpretation and preparation of the report

Unit II: Research problems and review of literature **(12 h)**

- Defining and selection of problem, necessity and techniques.
- Need of research review
- Sources of literature and search strategies
- Research reading and note taking
- Bibliography, webliography and literature citation

Unit III: Research design and hypothesis **(12 h)**

- Concepts: types of variables, hypothesis, control, treatment, experimental units etc.
- Types of research design: exploratory, descriptive and diagnostic, hypothesis-testing
- Basic principles of experimental designs
- Important experimental designs: Informal and formal
- Hypothesis: Concept, need, characterization, testing, decision rule, two-tailed and one-tailed test

Unit IV Data collection and analysis **(12 h)**

- Sampling: steps, types, criteria of section of technique, errors
- Measurement of scales and indices
- Methods and tools of data collection (primary, secondary),
- Case study, field study, survey
- Processing of data: editing, coding, classification and tabulation
- Statistics in research: central tendency, dispersion, skewness, measures of relation

Unit V: Interpretation, Discussion and report writing **(12 h)**

- Interpretation: meaning, importance, technique and precaution
- Structure and content of Discussion
- Numbering and caption of figures
- Report writing: steps, type, componentsand formatting
- Presentation of research : oral and research paper
- Plagiarism: Concept, prevalence, factors, strategies to tackle and detection

References:

1. Ramamurthy G.C. (2011). Research methodology, kogent learning solution.inc Dreamtech Press
2. Joshua O. Miluwi and Hina Rashid R.M (2015). Principle method and practices, Mangalam Publication
3. KrishnaswamyO.P., Reddy D. Obul(2010) Research Methodology and Statistical Analysis, Himalaya Publishing House **ISBN-10:** 8184885970
4. VenkatamunireddyR.(2011) Fundamental of research, Deep amd Deep publication
5. Wilson, P. D. K., Wilson, K., & Walker, J. (Eds.). (2000). Principles and techniques of practical biochemistry. Cambridge University Press.

6. Gurumani, N. (2019). Scientific thesis writing and paper presentation. MJP Publisher, Chennai
7. Gurumani, N. (2014). Research methodology for biological science, MJP Publisher, Chennai
8. Kothari, C. R. (2004). Research methodology: Methods and techniques. New Age International.
9. Malmfors, B., Garnsworthy, P., & Grossman, M. (2003). Writing and presenting scientific papers. Nottingham University Press.

M.Sc. (Microbiology): Semester-III
MB - 306: Agricultural microbiology

Hours: 60 **Credits: 4**

Course objectives:
- To introduce various attributes of microbial ecology and plant microbe interactions
- To learn mechanism of plant defence and its biocontrol
- To know recent approaches in agricultural microbiology

Course outcomes: On completion of this course, the student will be able to
- Understand microbial ecology and mechanism of plant microbe interactions
- Describe mechanism of plant and pathogen interactions and its biocontrol
- 3. Gain insight of recent approaches such as Rhizosphere engineering, GMO etc.

Unit I: Microbial ecology **(12 h)**
- Basic microbial ecology and its components
- Microbial interactions: positive and negative
- Microbial communities: concepts, elements and methods of analysis: CLPP, PLFA, DGCE, SSCP, ARDRA, FISH
- Methods to quantitative microbial ecology
- Indicators of soil health

Unit II: Microbial interactions with plant roots **(12 h)**
- Rhizosphere and its anatomy
- Mycorrhizae: VAM, OM, EM, Ectomycorrhiza
- Plant Growth Promoting Rhizobacteria (PGPR)
- Strategies for rhizosphere and mycorrhizae community study

Microbial interaction with aerial plant structure
- Phylloplane, Stems/ flowers, leaf buds
- Strategies for aerial plant structure study

Leguminous root nodules
- Nodulation process and mechanism of nitrogen fixation, *nif*operon
- Strategies to study infection process, root nodulation and N_2 fixation

Unit III: Pathogenic interactions with plants **(12 h)**
- Plant defense mechanisms (structural, biochemical, HR, SAR)

- Microbial pathogenicity mechanisms in virus, bacteria, fungal pathogens
- Genetic basis of plant-pathogen interactions
- Region-specific plant diseases (etiology, symptoms and control): Red rot of sugarcane, Sigatoka disease of banana, Powdery mildew, Smut and Rust

Unit IV: Microbial Bio control of plant disease (12 h)

- Methods of plant disease detection: traditional and innovative
- Plant disease control: general strategies and Principal of IDM
- Bio pesticides: BT, Siderophore and *Trichodermca,Pseudomonas*, NPV, *Beauveriabassiana*
- Bio control of post-harvest diseases
- Control of plant pathogens by genetic engineering

Unit V: Current approaches in agriculture microbiology (12 h)

- Integrated Plant Nutrition through biofertilizers
- Phytoremediation: Rhizodegradation
- Rhizosphere engineering
- Microbial reclamation of saline and sodic soils
- Genetically modified crops
- Microbiomes

References:

- Stanier RY, Ingraham JL, Wheelis ML and Painter PR (1993) General Microbiology, 5th edn., The McMillan Press Ltd., London (ISBN: 0-333-41768-2).
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- Press Inc., London (ISBN: 1-56022-328-6).

M.Sc. (Microbiology): Semester IV

MB - 401: Fermentation technology

Hours: 60 **Credits: 4**

Course Objectives:

- To introduce upstream processing in microbial fermentation and bioreactor operation
- To familiarize with recovery techniques used for fermentation products
- To introduce the microbial process adopted for production of various metabolites

Course Outcomes: On completion of this course, the student will be able to:

- Justify industrially relevant microbial products, their production and recovery process
- Get knowledge about bioreactor configuration
- Understand regulatory procedures required for final product.

Unit I: Upstream processing **(12 h)**

- Overview of typical microbial fermentation process (sterilization, inoculum, sampling, aeration, control system, cleaning)
- Microbial growth: kinetics, measurement
- Inoculum:
    - Criteria for good inoculum (purity, stage of growth, physiology, quantity, quality)
    - Acclimatization, seed media
    - Development of inoculum for following processes with one example: Yeast, Mycelial, Bacterial.
    - Aseptic inoculation of fermenter
- Media for industrial fermentation:
    - Types (synthetic and natural)
    - Formulation and Optimization
- Fermentation- kinetics of batch and continuous culture
- Strain improvement: mutation, recombination, Parasexual Cycle, Protoplast fussion and rDNA technique

Unit II: Bioreactor (Design and Application) and its operation **(12 h)**

- Design and construction materials of bioreactor
- Types of Bioreactor: continuous stirred tank, air lift, tower, fluidized bed, photobioreactor,
- Sterilization: Del factor, batch and continuous sterilizationand air(heat, filtration)
- Maintenance of aseptic condition
- Fermentation Process Parameters and Monitoring: aeration and agitation system, sterility, pH, temperature, foam, DO, Pressure and inlet& exit gas analysis. and process of automation
- Solid state fermentation: concept, bioreactor, advantages and disadvantage
- Scale up of fermentation:Major factors involved, Sale-up window, methods
- Mass and Heat transfer during fermentation

Unit III: Downstream processing **(12 h)**

- Biomass harvesting and removal of solid matter:

    - Criteria for choice of recovery
    - Methods: centrifugation, filtration, flocculation
- Cell disruption: mechanical,physical, chemical and enzymatic methods
- Product concentration : evaporation, liquid –liquid extraction, supercritical fluid extraction, ultrafiltration/membrane filtration, precipitation, three phase partitioning,
- Product purification and characterization: chromatography- adsorption, size exclusion, affinity, ion exchange, reverse phase, HPLC
- Formulation: Drying (spray drying, freeze drying)
- Biosafety: Handling infectious and Recombinant microorganism
- Production economics: cost (capital, operating), factors affecting

Unit IV: Microbial Products (12 h)

- Enzymes: Protease, Penicillin acylase
- Organic acids: Gluconic acid
- Amino acids: L glutamic acid
- Polysaccharides: Polysaccharides (Alginate and Hyaluronic acid)
- Others: Probiotics and Yoghurt,
- Antibiotics: Rifamycin
- Ethanol: 1 st, 2nd and 3rd generation
- Nucleotides: IMP, GMP
- Heterologous protein production with example

Unit V: Mammalian Cell Products (12 h)

- Media of animal cells: types (artificial, natural), general composition of complete media, physicochemical properties
- Cultured cell:
    - Characteristics, classification, cell proliferation and differentiation, measurement of growth parameters
    - Concept of senescence and apoptosis
- Primary culture: Techniques of disaggregation - mechanical and enzymatic
- Cell lines: types (finite and continuous), monoculture
- Scaling up of Animal Cell Culture
- Mammalian cell products: Monoclonal antibodies, Vaccines
- Heterologous protein: INF, tissue plasminogen activator (TPA), GMCSF

References:

- Stanbury, P.F., Whitaker A. and Hall, S.J. (2016) Principles of Fermentation Technology, 3rd Edition, Butterworth-Heinemann, Amsterdam, ISBN: 9780080999531
- Mukhopadhyay SN (2007) Process Biotechnology Fundamentals, 2nd edn., Viva Books, Mumbai, (ISBN: 81-7649-496-8).
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M.Sc. (Microbiology): Semester IV

MB-402: Applied molecular biology

Hours: 60 **Credits: 4**

Course Objectives:

- To learn about the various enzymes involved in and methods of rDNA Technology
- To know the concepts in microbial genomics and proteomics.
- To making aware of techniques in molecular biology and protein engineering

Course Outcomes: On completion of this course, the students will be able to:

- Understand tools of rDNA such as enzyme, vectors and basic ideas on methods of rDNA
- Know about genomics such as genome sequencing, mapping and editing.
- Understand the protein engineering with mapping and protein and protein interactions.

Unit I: Tools of molecular biology (or rDNA technology) (12 h)

- **Enzymes**: Restriction endonucleases and its types, DNA methylases, DNA polymerase, DNA ligases, Kinases, Phosphatases, topoisomerase
- **Cloning vectors**: Choice and its properties, Bacterial vectors: plasmid, Bacteriophage, Cosmids, Phagmids, BACs. Eukaryotic vectors: YACs, Ti, SV40
- **Cloning hosts**: Prokaryotic and eukaryotic hosts: properties

Unit II: Methods in rDNA technology (12 h)

- Vector mediated and chromosomal integration
- Genomic and cDNA library construction
- Gene transfer techniques: Transfection, Electroporation, Microinjection, Biolistic
- Screening, analysis and confirmation of rDNA

- Genetic methods
- Hybridization techniques - Dot Blot, Colony, Dip stick, Plaque
- Immunochemical methods
- Plus and minus screening, HRT and HART
- Analysis - Restriction mapping, Blotting techniques
- Confirmation by genetic marker and reporter genes

- Applications of genetic engineering

Unit III: Microbial Genomics (12 h)

- Concept of - Genome density, GC content, CPG Islands, Isochores, codon usage bias, cDNAs and ESTs, Contigs, epigenomics
- Structural, Functional, Application and Comparative Genomics:
  - Methods for whole genome sequencing, gene annotation o Gene and SNP identification
  - Genome mapping (Conjugation, Recombination and complementation) and map integration
  - Genome editing using CRISPR-cas system
- Concept of Metagenomics

Unit IV: Protein Engineering and Proteomics (12 h)

- Protein identification and Expression Mapping: 2D-gel electrophoresis, Mass Spectrophotometry and isotope labelling
- Protein-ligand docking
- Experimental approach to Protein-Protein interaction mapping:
  - Yeast and Bacterial 2-hybrid systems
  - Protein-ligand interactions
  - Protein fragment complement assays
- Protein arrays and chips: Antibody and peptide arrays

Unit V: Techniques in Molecular biology (12 h)

- DNA Sequencing : Sanger, Maxum Gilbert and high throughput [Polony, 454 pyrosequencing, Illumina (Solexa), Massively parallel signature sequencing (MPSS), SOLiD, Ion Torrent semiconductor, single molecule, Single molecule real time (SMRT)]
- PCR: Basics, Reverse transcriptase PCR, Real time PCR, Applications
- Analysis of polymorphism: RFLP, RAPD, AFLP, SSCP, DGGE
- Analysis of gene expression : SAGE, Microarray

References:

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M.Sc. (Microbiology): Semester IV

MB- 403: Methods in Biostatistics and Bioinformatics (Practical Course)

Hours: 120 **Credits: 8**

Course objectives:

- To impart training about elementary aspects of statistics used in microbiology
- To introduce variety of computational methods for predicting functionalbehaviour of biological system
- To analyse the output data to predict a biologically relevant function

Course outcomes: On completion of this course, the student will be able to:

- Able to analyse experimental data with central tendency, its dispersion and presentation graphically
- Access biological databases, interpret structural aspects
- Familiar with statistical and bioinformatics software

1. Calculate mean, median, mode, range, variance, standard deviation, standard error, confidence interval using MS-Excel/suitable software
2. Plot straight Line (Linear Least squares) using LINEST Function of MS-Excel/ suitable software
3. Plot - line, scatter graphs, bar graphs, error bars using MS-Excel/ suitable software
4. Determine: linear regression, Correlation and their coefficients using MS-Excel/ suitable software
5. Compute F-test (paired and unpaired), t-test, using MS-Excel/ suitable software
6. Compute ANOVA, Chi 2-test using MS-Excel/ suitable software
7. Demonstration of multivariate analysis of process parameters using statistical tools.
8. Exploring Biological databases - Genbank& Protein Data Bank
9. Structural predication of protein using ExPASy software
10. Primary and tertiary structure analysis of protein/ DNA using BLAST
11. Multiple sequence alignments using Clustal W
12. Phylogenetic tree analysis using MEGA
13. Primer designing using biological software

References:

- Bailey NTJ (1959) Statistical methods in Biology, ELBS and The English Universities Press Ltd., UK.
- Irfan Ali Khan and AtiyaKhanum (2004) Fundamentals of biostatistics, Ukaaz Publication, Hydrabad.
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- Mount DW (2001) Bioinformatics: sequence and genome analysis. Cold Spring Harbor Laboratory Press, New York.

M.Sc. (Microbiology): Semester IV

MB-404: Laboratory course (Project Dissertation)

Hours: 120 **Credits: 8**

Course Objectives:

- To give exposure to the students to research methodology
- To introduce criteria for selection of research topic, plan and perform experiments, collect and analyse the data
- To foster independent thinking

Course Outcomes: On completion of this course, the student will be able to:

- Carry out comprehensive survey of literature and comprehend a problem based on review
- Plan experimental framework for research and present the work in written format.
- Present the research with ICT tools and face viva voce

The project is allotted during the Forth semester. The students will get an opportunity to become a part of ongoing research activities in the respective College. The student will explore and gain experience in different sectors of biotechnology viz agriculture, food, medicine and pharmaceutical. The students will acquire skill to write, compile and analyze data, and present the detailed technical/scientific report. At the end of successful project semester training, potentially the students become employable in the industries/organizations.

It is expected that the students will design experiments and collect experimental data to deduce conclusions. At the end they will submit a detailed thesis for evaluation. The students should be introduced to research methodology in the beginning through few lectures.
The approach towards the execution of project should be as follows:

1. Selection of topic relevant to priority areas of biotechnology.
2. Collection of literature on the topic of research from libraries, internet, on-line journals, Planning of research experiments
3. Performing the experiments with scientific and statistical acceptability.
4. Presentation of observations and results.
5. Interpretation of results and drawing important conclusions.
6. Discussion of obtained results with respect to literature reports.
7. Writing monthly progress report
8. Preparation of report (Dissertation) containing introduction, materials and methods, results and discussion, conclusions, bibliography and submission of at least 3 copies (1 copy retained in the department and after examination submitted to Library, 1 copy submitted to the guide and 1 copy kept with the candidate).
9. Presentation of research data during university examination and submission of project dissertation in a bound form.

1. **Internal examination (40 marks):** Components of continuous internal assessment Submission of monthly progress report and signed by supervisor (at least 4 reports) (2 marks per report = 8 marks), Literature collected, experiment planning and design (10 marks), Experiments conducted (10 marks), outcome of the experiments and viva (8 marks) and regular attendance (4 marks) recorded: Research Supervisor

2. **External examination (60 marks)** and Components of external assessment:
   Subject matter (5 marks), Review of literature (10 marks), Writing of dissertation submitted in bound form at the time of examination (Title page, Certificate, Plagiarism report, Main content: Abstract, Introduction, Literature, Materials and methods, results and discussion and conclusion with relevant references) (15 marks), Presentation structure (PPT format) (10 marks), Overall presentation reflecting contribution of work (5 marks), Response to questions (15 marks).
   References: Refer to the journals, reference books, abstracts etc. related to topic

M.Sc. (Microbiology): Semester IV

MB-405 : Biostatistics and bioinformatics

Hours: 60 **Credits: 4**

Course objectives:

- To impart understanding of elementary aspects of bio statistics
- To introduce overview of bioinformatics viz. data sequencing and mining
- To analyse the output data to predict a biologically relevant function

Course outcomes: On completion of this course, the student will be able to:

- Practice biostatistics for interpretation of experimental data
- Understand fundamentals of database bioinformatics

- Access information from databases and interpret phylogenetic tree to gain insight into evolutionary path

Unit I: Sampling and central tendency (theoretical concepts) (12 h)

- Applications of bio statistics to biological database
- **Concepts**: population, sample, probability, central tendency, dispersion, variance
- **Sampling** types: simple random, stratified, systematic, cluster
- **Data**: collection (primary and secondary), classification and tabulation of data
- Graphical techniques: one dimensional, two dimensional, pictograms
- Graphical presentation of data: histogram, frequency polygon, frequency curve, ogive curve
- **Measures of Central tendency**: Mean, Mode, Median
- **Measures of dispersion:** range, mean deviation, standard deviation, coefficient of variation, skewness

Unit II: Correlation, Regression, probability, ANOVA (theoretical concepts) (12 h)

- **Correlation**: definition, types, methods of measuring
- **Regression**: definition, equation, coefficient,Simple linear regression,
- **Analysis of Variance:** One and two way analysis of variance, Intro experiments, Multivariate statistical analysis
- **Probability:** Definition, concepts: experiment, event (simple, compound, mutually exclusive, independent, dependent)
- **Discrete distributions**: Bernoulli, Binomial, Poisson. Continuous: Normal, Exponential, sigma limits and probability coverage.
- Concepts: Hypothesis testing, t test, F test, Chi square test, Design of experiments, multivariate statistics

Unit-III: Overview of Bioinformatics (12 h)

- Concept of Bioinformatics: definition, scope, application and limitations
- Introduction to biological databases
    - Primary (GenBank, EMBL, SWISS-PROT)
    - Secondary (PROSITE, pfam)
    - Specialized (GenBank EST and Microarray Gene Expression)
    - UNIPROT database
- Structural databases: PDB, MMDB
- Sequence retrieval system - SRS, ENTREZ, Expasy
- Bioinformatics servers in India

Unit-IV: Sequence analysis and phylogeny (12 h)

- Sequence analysis: Concept of % Identity, Twilight zone and mid night zone
- Concept of homology, analogy, othology and paralogy
- Sequence alignment: pair wise (local, global) and multiple sequence (Clustal W editing and interpretation)
- Alignment Algorithms: Dot Matrix and Dynamic Programming, Gap Penalties
- Introduction to scoring matrices-PAM and BLOSSUM
- Database similarity searching: BLAST(Variants and statistical Significance) FASTA (statistical Significance)

- Phylogenetic analysis: Definition, Choice of Molecular Markers
- Phylogenetic tree: Terminology, construction methods (maximum Parsimony, Maximum Likelihood), Evaluation (Bootstrapping and Jackknifing, Phylogenetic Programs).

Unit –V: Mining and Data Visualization (12 h)

- Protein structure prediction
    - Secondary structure prediction: Globular and Transmembrane
    - 3D Structure prediction: X ray Crystallography, Nuclear Magnetic Resonance Spectroscopy
- Classification of protein Structure: SCOP
- Drug discovery: Target selection, Prediction of a lead compound, Molecular modeling
- Software for Data Visualization: CN3D,Rasmol, Chimera, SWISS PDB Viewer, EXPASY
- Application of programming Language in Biology: Bioperl, Biojava and R Programing

References:

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M.Sc. (Microbiology): Semester IV

MB 406: Entrepreneurship in microbiology

Hours: 60 **Credits: 4**

Course objectives:

- To impart understanding of business management and entrepreneurship
- To introduce skills required for bio-entrepreneurship
- To understand basics of business ethics, safety and IPR

Course outcomes: On completion of this course, the student will be able to:

- Understand fundamental business, management, marketing, operation and HRM
- Able to develop skill set required for entrepreneurship in microbiology
- Address the issues related to business ethics and understand concept of safety and IPR

Unit I: Introduction to Business and management (12 h)

- Management
  - Definition, concept, importance
  - Levels (individual, first, middle and top),
  - Functions (planning, organization, staffing, directing, controlling)
  - Skills (technical, human, conceptual, diagnostics, communication, political)
  - Organization types, coordination, control and decision making
  - Strategic management: characteristics, importance, SWOT analysis
- Production and operation
  - Objectives, functions
  - Planning and control, Total quality management, scale forecasting
- Marketing, Marketing research- concept and techniques
- Core concept of Market: Identification and evaluation of market potential of various bio-entrepreneur sectors
- Financial management: objective, method and importance
- Concept of capital budgeting and working capital
- Concept of human research management and planning

Unit II: Business development and entrepreneurship (12 h)

- Conceptual framework and characteristics for being an entrepreneur in microbiology.
- Factors affecting microbiology/ biotechnology business:
  - Finance, infrastructure, equipment, manpower, resources, project location, end product, quality issues, etc.
- Role of government and schemes
- Financial institutions in fostering bioentrepreneurship.

Unit III: Business management, skills in bio-entrepreneurship (12 h)

- Personality and attitude, organizational behavior, leadership
- Principles of effective communication. Body language, public speaking, presentations,
- Business proposal writing.
- Communication aid and application of technology
- Case studies of successful and unsuccessful bio-entrepreneurship.

Unit IV: Ethics, regulation and safety (12 h)

- Business ethics: meaning, principals (Integrity, Honesty, Respect and Concern, Fairness), Influential factors, ethical dilemmas in biotech industry
- Regulations related aspects and laws for industry
- Current regulatory bodies
- Translational development
- Human clinical testing phases
- Biologics License Application (BLA)
- Safety and precautions
    - Biological: microbes, sterilization, documentation, disposal, storage
    - Chemical: MSDS, cautions
    - Personal: flame, solvent, apron, mask, eyes etc.
- Biosafety: leverls (BL 1, 2, 3, 4), GMOs and risks

Unit V: Intellectual property and IPR (12 h)

- IPR- Introduction, Forms of IPR: patent, copyrights, geographical indications, trademarks, trade secret, Industrial designs and Patent law
- Patentable and non-patentable items
- Legislations covering IPR's in India
- Overview of patent system: Steps in filing patent, provisional and complete specification
- Valuation of patent and business concerns
- Protection of biotechnological inventions: biological materials such as life forms, transgenic materials, genetic resources
- Patent regulatory bodies at National and International level
- Overview of GATT, role of WTO and TRIP
- Acts: Indian patent act 2005, Indian biodiversity act
- Protection of plant varieties and farmer's rights

References:

- Mehta, S. S. (2008) Commercializing successful biomedical technologies: basic principles for the development of drugs, diagnostics and devices. Cambridge University Press.
- Patzelt, H., & Brenner, T. (Eds.). (2008) Handbook of bioentrepreneurship (Vol. 4). Springer Science & Business Media.
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- Modi H.A. (2008) Fermentation Technology (Vol 1 & 2), Pointer Publishers, Jaipur.

Additional Instructions:

- Theory examination (60 marks) will be of three hours duration for each theory course. There shall be 5 questions each carrying equal marks (12 marks each). The pattern of question papers shall be:
  - Question 1 (12 marks): 6 sub-questions, each of 3 marks; answerable in brief and based on entire syllabus, attempt any 4 out of 6 questions.
  - Question 2, 3 and 4 (12 marks each): based from Unit I & II, III & IV, V & any Unit I to IV, respectively, each question has 3 sub-questions of 6 marks each and answer only 2 subquestions from each Q2, Q3, and Q4.
  - Question 5 (12 marks): answer only 3 out of 5 in brief, based from all 5 units, Each 4 marks.
- Internal examination (40 marks each semester): Internal assessment of the student by respective teacher will be comprehensive and continuous, based on written test. The written test shall comprise of both objective / subjective type questions.
- Practical Examination: Practical examination shall be conducted by the college at the end of the semester. Practical examination will be of minimum 5-6 hours duration and shall be conducted as per schedule (10 am to 5 pm on schedule date or can be scheduled 10 am - 1pm/ 2-5 pm for 2 consecutive days) in case of microbiology practicals where incubation condition, allied aspect are essential. There shall be 5 marks for laboratory log book and well written journal, 10 marks for viva voce and minimum three experiments (major and minor). Certified journal is compulsory to appear for practical examination. There shall be one expert and two examiners (external and internal) per batch for the practical examination.

Skills imparted:

The curriculum is designed to instill basic and applied knowledge of the subject to the students. One of the major objectives considered during designing is to make technically educated human resource. Basic microbiology, molecular biology, microbial physiology may help to find out unseen facts in various environmental, agriculture, food and pharmaceutical sectors. The subjects like genetic engineering, applied microbiology, microbial biochemistry, pharmaceutical microbiology, fermentation technology and biochemical techniques are designed to impart theoretical and practical knowledge of modern scientific advances in the field. Further to enhance skillful human resource with precision, the course like biostatistics and bioinformatics are included. The subject like Microbial biotechnology would give not only the practical knowledge of industry and industrial processes but also make aware the students with the global environmental problems like pollutions, contamination and bioremediation. Practical courses are based on theory courses and are designed to improve research oriented skills of students.

Job opportunity:

The designed curriculum offers job opportunities in various sectors like,

- Pharmaceutical industry: Clinical, medicine, vaccine, QC division
- Biotech industry: Recombinant product, QC, QA
- Agrochemical and pesticide industry
- Chemical industry: synthesis, testing
- Environmental protection industry and Agencies
- Research leading to Ph. D. degree
- Self entrepreneurship

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