

M.Sc. (Microbiology) Semester-III

Paper I-Molecular Biology and Genetic Engineering

UNIT-I

10 lectures

Essential enzymes used in r-DNA technology.
Restriction digestion, ligation and transformation.
Cloning vectors- Plasmids, phages and cosmids.
Animal virus derived vectors (SV 40, Vaccinia), plant based vectors (Ti, Ri vectors)
Artificial chromosomes as vectors (YAC and BAC vectors)
Cloning strategies-Cloning and selection of individual genes, gene libraries - cDNA and genomic libraries.

UNIT-II

10 lectures

Specialized cloning strategies –
Expression vectors: basic features of expression vectors, promoters used in expression vectors, pMal, GST, pET based vectors
Cassettes and gene fusions
Fusion vectors-Translational and transcriptional fusion vectors
Recombinants protein purification-advantages of fusion proteins, fusion proteins tags-His-tag, GST-tag, MBP-tag, methods involved in recombinants protein purification

UNIT-III

10 lectures

DNA sequencing methods-Sanger method, Maxam and Gilbert method, Thermal cycle sequencing, Pyrosequencing, Automated DNA sequencing. Assembly of contiguous DNA sequence
Gene amplification - PCR-principle, types and applications.
DNA microarray technique.

UNIT-IV

10 lectures

Expression of cloned DNA – Expression in heterologous system.
Study of the transcript of a cloned gene.
Hybridization techniques-Colony hybridization, plaque hybridization, Southern, Northern, Western and Southwestern blotting, in situ hybridizations.
Modification of cloned DNA – Site directed mutagenesis, transposon mutagenesis.

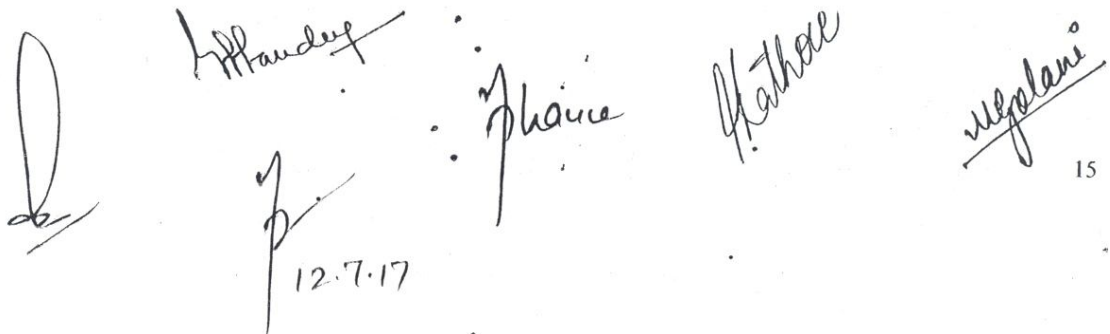
UNIT-V

10 lectures

Applications of r-DNA technology-Requirement and production of recombinant molecules in pharmaceutical, health, agricultural and industrial sectors and research laboratories.
Transgenic animals, *Agrobacterium* mediated transformation, Bt cotton, Gene Therapy.
Ethical and safety issues associated with recombinant DNA technology, IPR and patenting.

REFERENCES

1. Genomes- Brown TA, 2nd edition (Oxford: Wiley-Liss)
2. Principles of Gene Manipulation - Old RW & Primrose SB, 5th edition (Blackwell Scientific Publications)
3. Gene Cloning & DNA Analysis – Brown TA, 6th edition (Wiley Blackwell)
4. Molecular Biotechnology- Glick BR, Pasternak JJ & Patten CL, 4th edition (ASM Press, Washington)
5. Principles of Gene Manipulation and Genomics-Primrose SB & Tyman RM, 7th edition (Blackwell Publishing)
6. Genetic Engineering-Rastogi S & Pathak N, 2009 (Oxford University Press)
7. Genetic Engineering: Principles and Practice-Mitra S, 2nd edition (McGraw Hill Education, New Delhi)
8. Molecular Biology of the Gene-Watson JD, Gann A et al., 7th edition (Cold Spring Harbor Laboratory Press, New York)
9. Current Protocols in Molecular Biology- Ausbel FM *et. al.*, 2003 (John Wiley & Sons)
10. Molecular Cloning Vol. I-III - Sambrook and Russel, 2001 (CSH press)
11. Genome analysis (Four Volumes)- Paterson AH, Brubaker CL, Wendel JF, 1993 (Springer, CSH Press)
12. DNA Microarrays: A Practical Approach-Schena M (Oxford University Press)



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Paper II-Medical Microbiology

10 lectures

UNIT-I

Epidemiological studies of infectious diseases-
Reservoirs and sources of diseases.
Infection and its types, transmission of infections.
Types of diseases-epidemic, endemic, pandemic, sporadic
Preventive and control measures for diseases, Hospital acquired infections and their prevention
Epidemiological Methods – Descriptive, Analytical and Experimental Epidemiology.
Introduction to Centers for Disease Control and Prevention (CDC) & National Centre for Disease Control (NCDC)

10 lectures

UNIT-II

Normal microbial flora of human body
Classification and identification of medically important microorganisms
Opportunistic pathogens and true pathogens
Attributes predisposing to microbial pathogenicity- virulence: attenuation and exhalation, infecting dose
Microbial pathogenicity - Mechanism and factors involved in establishment and spreading of infection, adhesion, invasiveness, toxigenicity.

10 lectures

UNIT-III

Recent advances in medical microbiology-
Multidrug resistance: Antibiotics-types & mode of action. Types, biochemical mechanisms and development of multidrug resistance. Guidelines for rational use of antibiotics: Multidrug-resistant organisms- Methicillin-Resistant *Staphylococcus aureus* (MRSA), Extended Spectrum β -lactamase (ESBL) producing Gram-negative bacteria, MDR & XDR tuberculosis, Carbapenem resistant *Enterobacteriaceae* (CRE).
Introduction to emerging diseases- Dengue hemorrhagic fever, Swine flu, Chicken guenica, Ebola, SARS.
Overview and current status of anti HIV, anti malaria and anti tuberculosis treatment.

10 lectures

UNIT IV

Etiology, clinical features, pathogenesis, laboratory diagnosis, transmission, prevention & control of diseases caused by-
Gram positive cocci - *Staphylococcus aureus*, *Streptococcus* species.
Gram positive bacilli - *Clostridium* species.
Gram negative cocci- *Neisseria* species
Gram negative bacilli - *E.coli*, *Salmonella* species
Acid Fast Bacteria – *Mycobacterium tuberculosis*

10 lectures

UNIT-V

Etiology, clinical features, pathogenesis, laboratory diagnosis, transmission, prevention & control of diseases caused by-
Actinomycetes- *Actinomyces israelii*.
Spirochaetes- *Treponema pallidum*.
Rickettsiae- *Rickettsia* species
Chlamydiae- *Chlamydia* species.
Fungi: *Microsporium*, *Trichophyton*, *Candida albicans*.
Virus- Hepatitis virus, HIV, Polio virus
Protozoa- *Plasmodium* species, *Entamoeba histolytica*.

REFERENCES

1. Medical Microbiology - Mackie TJ & McCartney JE (Vol. 1-Microbial Infection & Vol. 2-Practical Medical Microbiology), 13th edition (Churchill Livingstone, London)
2. Text book of Microbiology-Ananthnarayan R & Panikar CKJ, 8th edition, (University Press Pvt Ltd, Hyderabad)
3. Basic Medical Microbiology-Boyd RF, 5th edition (Little Brown, Boston)
4. Bailey and Scott's Diagnostic Microbiology-Forbes BA, Sahm DF & Weissfeld AS, 12th edition (Mosby, Elsevier)
5. Microbiology in Clinical Practice – Shanson DC, 1st edition (Wright, London)
6. Biochemistry of Antimicrobial Action – Franklin TJ, Snow GA & Hall, 4th edition (Chapman and Hall Ltd.)
7. Epidemiology and Infections – Smith CEG, 1st Edition (Meadowfield Press Ltd, England)
8. Bacterial Disease Mechanism-An Introduction to Cellular Microbiology - Wilson M, McNab R & Henderson B, 2002 (Cambridge University Press)

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M. Sc. (Microbiology) Semester- III
Paper III-Biostatistics and Bioinformatics

UNIT-I

10 lectures

Basic definitions and applications of statistics in biological research.

Sampling: Representative sample, sample size, sampling bias and sampling techniques.

Data collection and representation: Types of data, methods of collection of primary and secondary data. Methods of data representation: tabular, diagrammatic graphical representation by bar diagram, histogram, polygon and pie diagram.

Measures of central tendency: Mean, Median, Mode.

Measures of variability: Standard deviation, standard error, range, mean deviation and coefficient of variation.

UNIT-II

10 lectures

Test for significance: Small sample tests (Chi-square test, F test), large sample test (Z test)

Analysis of variance (ANOVA): analysis of variance in one-way and two way classification.

Correlation and Regression: Positive and negative correlation and calculation of Karl-Pearson's co-efficient of correlation. Linear regression and regression equation.

Introduction to probability theory and distributions (concept without deviation).

UNIT-III

10 lectures

Introduction to bioinformatics-History, aim and scope of bioinformatics.

Database concept, DBMS.

Classification of biological databases and their functions-

Nucleotide sequence database: EMBL, GenBank, DDBJ

Protein sequence database: SWISS-PROT, PIR

Nucleic acid and Protein structure databases – NDB, PDB

Protein structure classification databases (SCOP, CATH)

Genome databases and composite database (NCBI)

Specialized databases (ESTs, EXPASY, Prosite, Pfam)

UNIT-IV

10 lectures

Basic concepts of sequence comparison, sequence identity, similarity and homology.

Scoring/substitution matrices (PAM, BLOSUM).

Sequence database searching tools (BLAST, FASTA)-basic knowledge of their variants and statistical significance.

Pair wise Local and Global sequence alignment algorithms (Needleman & Wunsch, Smith & Waterman).

Multiple sequence alignment.

UNIT - V

10 lectures

Studying Open reading frames (ORFs), Motifs, Domains, Patterns, Profiles and their importance.

Phylogenetic analysis-Basic concepts of phylogenetic analysis, rooted/uprooted trees, approaches for phylogenetic tree construction (UPGMA, Neighbour joining methods). Introduction to Operational taxonomic units (OTU).

REFERENCES

1. Primer of Biostatistics, Glantz S, 7th edition (McCraw Hill Companies)
2. Statistics for Biology-Bishop ON, 3rd edition (Longman Publishing Group)
3. Statistics in Biology-Bliss CI, (McGraw Hill Companies)
4. Practical Statistics for Experimental Biologists-Wardlaw AC, 2nd edition (John Wiley & Sons)
5. Statistical Methods in Biology-Bailey NTJ, 1st edition (English Universities Press)
6. Statistics for Biologists-Campbell RC, 3rd edition (Cambridge University Press, UK)
7. Fundamentals of Biostatistics- Irfan AK & Khanum A, 2014 (Ukaaz Publications, Hyderabad)
8. Biostatistics – Arora PN, Malhan PK, 1st edition (Himalaya Publishing House, Mumbai)
9. Bioinformatics: A Practical Guide to the Analysis of Genes and Proteins (Methods of Biochemical Analysis)-Baxevanis AD, Ouellette BFF, 1st edition (John Wiley & Sons)
10. Bioinformatics: Sequence, Structure, and Databanks : A Practical Approach-Higgins D, Taylor W, 1st edition (Oxford University Press)
11. Bioinformatics: A Practical Guide to the Analysis of Genes and Proteins-Baxevanis AD, Ouellette BFF, Volume 43, 2nd edition (John Wiley & Sons)
12. Bioinformatics Methods & Protocols (Methods in Molecular Biology)-Misener S, Krawetz SA, 1999th edition (Humana Press) N, Malhan PK, 1st edition (Himalaya Publishing House, Mumbai)

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Paper IV-Applied Microbiology

Unit I

10 lectures

Production of biofertilizers and bioinsecticides-

Production and methods of application of: *Rhizobium* biofertilizer, *Azotobacter* biofertilizer, *Azospirillum* biofertilizer, *Azolla* & Blue-Green Algae biofertilizer, phosphate solubilizing biofertilizer.
Quality control of biofertilizers as per FCO (Fertilizer Control Order)-method of analysis, standards of biofertilizers.

Production of Bioinsecticides : Candidate microorganisms, production, safety, advantages and disadvantages of bioinsecticides.

Unit II

10 lectures

Biofuel Production-

Biogas-The substrate digester and the microorganisms in the process of biogas production (biomethanation).
Bioethanol-Production of bioethanol from sugar, molasses, starch and cellulosic materials. Ethanol recovery.
Biohydrogen-Microbial production of hydrogen gas.
Microbial production of biodiesel from hydrocarbons.
Algae as biofuel.

Unit III

10 lectures

Bioremediation and Biosensors-

Microbiology of degradation of xenobiotics in the environment.

Strategies and techniques of bioremediation: *in situ* and *ex situ*.

Approaches to bioremediation- intrinsic bioremediation, biostimulation, bioaugmentation.

Bioremediation of oil spills, metals, lignins and hazardous wastes. Application of GMO in bioremediation.

Biosensors- definition, components of biosensors, principle of operation.

Methods of biomaterial and sensor coupling.

Types of biosensors. Applications of biosensors.

Unit IV

10 lectures

Biobleaching and Petroleum Microbiology-

Microbes and mineral recovery: General methods of biobleaching, biobleaching of copper, gold and uranium from low grade ores.

Petroleum microbiology-Microbial Enhanced Oil Recovery (MEOR), detrimental activity of microbes in petroleum industry.

Unit V

10 lectures

Bioplastics and Biosurfactants-

Definition of bioplastics, types of bioplastics, genetically modified bioplastics.

PHA-properties, types, chemical structure, PHA producing microbes.

Modern trends in microbial production of bioplastics.

Applications of bioplastics, biodegradability of bioplastics, advantages and disadvantages of bioplastics.

Biosurfactants- microbial production and applications.

REFERENCES

1. Microbial Biotechnology: Fundamentals of Applied Microbiology -Glazer AN and Nikaido H, 2nd edition, Cambridge University Press.
2. A Textbook of Basic and Applied Microbiology-Aneja KR, New Age International.
3. Biofertilizers- Somani LL ,Bhandari SC , Saxena SN , Vyas KK, Scientific Publishers , Jodhpur.
4. Biofertilizers in agriculture and forestry- Subba Rao NS (Oxford and IBH, Publishing Co. New Delhi)
5. Bioremediation- Baker KH and Herson DS, Mc Graw Hill, New York
6. Biodegradation and Bioremediation-Alexander, M, 2nd edition, Academic Press.
7. Textbook of Environmental Biotechnology-Mohapatra PK, I K International Publishing House Pvt. Ltd
8. Microbial Mineral Recovery-Ehrlich HL & Bierley CL , McGraw-Hill

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M.Sc. (MICROBIOLOGY) - SEMESTER III

LAB COURSE I

Paper I-Molecular Biology and Genetic Engineering

Paper II-Medical Microbiology

Suggested List of Practicals

1. Extraction of plasmid DNA and its analysis using agarose gel electrophoresis
2. Preparation of competent cells and transformation of DNA by using CaCl_2
3. Determination of molecular size of DNA fragments
4. Restriction digestion of DNA samples using restriction endonucleases
5. DNA fingerprinting technique- Restriction Fragment Length Polymorphism-RFLP
6. To perform ligation of DNA fragments
7. DNA purification from electrophoresed agarose gel (electro elution/silica based technique , etc.)
8. DNA amplification by Polymerase Chain Reaction (Demonstration)
9. DNA fingerprinting technique- Random Amplified Polymorphic DNA -RAPD (Demonstration)
10. Blotting techniques (Demonstration)
11. Isolation, biochemical characterization and identification of medically important bacteria-
 - *E. coli*, *Klebsiella pneumoniae*, *Salmonella typhi*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus sp.*
12. Determination of antibiotic susceptibility pattern of pathogenic microbes
13. Study of synergistic and additive effect of antibiotics
14. Isolation and identification of resident normal flora from skin/throat
15. Effect of disinfectants on microflora of skin
16. Haematology : RBC Count, Total WBC Count, Differential WBC Count, Haemoglobin estimation
17. Laboratory analysis of urine-physical, chemical, microscopic and bacteriological analysis

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M. Sc. (MICROBIOLOGY) - SEMESTER III

LAB COURSE II

Paper III-Biostatistics and Bioinformatics

Paper IV-Applied Microbiology

Suggested List of Practicals

1. Diagrammatic and graphical presentation of statistical data using MS Excel
2. Use of in-built statistical functions of MS Excel for computations of statistical averages, central tendency and standard deviation
3. Calculation of standard deviation
4. Calculation of standard error
5. Application of tests of significance
6. Introduction to NCBI and its database
7. Variants of BLAST and FASTA
8. Sequence manipulation suite
9. Pairwise sequence alignment
 - i. Global pairwise alignment using Needleman-Wunsch Algorithm based ALIGN EMBOSS tool
 - ii. Local pairwise alignment using Smith Waterman Algorithm based ALIGN EMBOSS tool
10. Multiple sequence alignment
11. Isolation of *Rhizobium* from root nodules
12. Isolation of *Azotobacter* from soil
13. Isolation of *Azospirillum* from soil
14. Isolation and characterization of phosphate solubilisers from soil
15. Isolation and characterization of PHA producing bacteria
16. Isolation and characterization of biosurfactant producing bacteria
17. Isolation and characterization of lignin degrading microorganisms
18. Isolation and characterization of dye degrading microorganisms from industrial effluents

A. P. Hande



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