

**MASTER OF SCIENCE  
in  
BIOTECHNOLOGY**

**TWO-YEAR FULL-TIME PROGRAMME  
(Semester wise Choice Based Credit System)**

***Curriculum and Syllabus***

**ALLAHABAD STATE UNIVERSITY,  
CPI campus, Mahatma Gandhi Road, Civil lines,  
ALLAHABAD– 211001 (U.P.)**



**MASTER OF SCIENCE  
IN  
(BIOTECHNOLOGY)  
TWO-YEAR FULL-TIME PROGRAMME**

**AFFILIATION**

The proposed programme shall be governed by Allahabad State University, Allahabad- 273009.

**PROGRAMME STRUCTURE**

The M.Sc. (Biotechnology) Programme is divided into two Parts as under. Each part will consist of two Semesters as given below. Semester-I, II and III has four core courses (three theory courses and one practical) and two elective courses (students can opt for any one elective course for the semester I, II and III). Semester -IV (Final Year) comprises of two core courses namely Dissertation and Seminar. The detail syllabus structure of the MSc (Biotechnology) programme is:

Course Number	Course Title	Type Core/Elective
<b>SEMESTER-I</b>		
MBT-C-101	Microbiology	Core
MBT-C-102	Biochemistry and Biophysics	Core
MBT-C-103	Cell and Molecular Biology	Core
MBT-E-101	Genomics & Proteomics	Elective
MBT-E-102	Bioentrepreneurship	Elective
MBT-C-104	Practical based on paper MBT-C-101, MBT-C-102&MBT-C-103	Core
<b>SEMESTER-II</b>		
MBT-C-201	Recombinant DNA Technology	Core
MBT-C-202	Immunology and Immunotechnology	Core
MBT-C-203	Bioinformatics and Biostatistics	Core
MBT-E-201	IPR and Biosafety Issues	Elective
MBT-E-202	Molecular breeding	Elective
MBT-C-203	Practical based on paper MBT-C-201, MBT-C-202& MBT-C-203	Core
<b>SEMESTER-III</b>		
MBT-C-301	Fermentation Technology and Downstream Processing	Core
MBT-C-302	Environmental Biotechnology	Core
MBT-C-303	Enzymology and Enzyme Technology	Core
MBT-E-301	Plant Tissue Culture and Transgenic Technology	Elective
MBT-E-302	Animal Cell Culture Technology	Elective
MBT-C-303	Practical based on paper MBT-C-301, MBT-C-302& MBT-C-303	Core
<b>SEMESTER-IV</b>		
MBT-C-401	Dissertations	Core
MBT-C-402	Seminar	Core



## **M.Sc (First Year) SEMESTER-I**

### **MICROBIOLOGY (MBT-C-101) THEORY**

#### **Unit-1:**

Microbial diversity and systematics, Modern approaches to bacterial taxonomy, polyphasic classification, General characteristics of primary domains and of taxonomic groups belonging to Bacteria, Archaea and Eukarya, Nomenclature and outline of bacterial classification as per Bergey's Manual, Accessing microbial diversity using molecular methods such as Denaturing Gradient Gel Electrophoresis (DGGE), Temperature Gradient Gel Electrophoresis (TGGE), Amplified rRNA restriction analysis, terminal Restriction Fragment Length Polymorphism (T-RFLP), 16S rDNA sequencing, metagenomics.

#### **Unit-2:**

Methods in Microbiology: Theory and practice of sterilization, Pure culture techniques, Principles of microbial nutrition, Construction of culture media, Enrichment culture techniques, Isolation and culture of aerobic and anaerobic bacteria, Culture collection, preservation and maintenance of microbial cultures.

#### **Unit-3:**

Metabolic Diversity among Microorganism: Microbial Nutrition: nutritional types and modes of nutrition in bacteria, Extremophiles. Microbial growth: The definition of growth, mathematical expression of growth, growth curve, measurement of growth and growth yields; Synchronous growth and Continuous culture.

#### **Unit-4:**

Chemotherapy / Antibiotics Antimicrobial agents; Antibiotics: Penicillins and Cephalosporins and Broad- spectrum antibiotics, sulfa drugs, Antifungal antibiotics, Mode of action, Molecular mechanism of drug resistance. Bacterial Genetic System: Transformation, Conjugation, Transduction, Recombination, bacterial genetic map with reference to *E. coli*.

#### **Books Recommended:**

- ❖ Brock Biology of Microorganisms, 9<sup>th</sup> Edition. by *Michael T. Madigan, John M. Martinko, Jack Parker*. Prentice Hall, Inc.
- ❖ Microbiology, 4<sup>th</sup> Edition. by *Lansing M. Prescott, John P. Harley, Donald A. Klein*. WCB McGraw Hill.
- ❖ General Microbiology, 5<sup>th</sup> Edition by *Roger Y. Stanier, John L. Ingraham, Mark L. Wheelis, Page R. Painter*, Macmillan Press Limited.
- ❖ Microbiology: Principles and Explorations, 5<sup>th</sup> Edition. by *Jacquelyn G. Black*, John Wiley & Son, Inc.



# BIOCHEMISTRY AND BIOPHYSICS

## (MBT-C-102)

### THEORY

#### Unit-1:

Amino acids and proteins : Classification, structure and properties of amino acids; primary, secondary, tertiary, quaternary and domain structure of proteins, forces stabilizing protein structure, Ramachandran plot, DNA-protein and protein-protein interactions, protein folding, protein misfolding and related diseases; protein sequencing. Nucleic acids : Primary and secondary structure of nucleic acids, Watson-Crick model of DNA, structural polymorphism of DNA and RNA, three dimensional structure of RNA, biosynthesis of purines and pyrimidines.

#### Unit-2:

Carbohydrates: Classification and structure of carbohydrates, polysaccharides, glycoproteins and peptidoglycans, glycolysis, TCA cycle, oxidative phosphorylation, glycogen synthesis and breakdown, gluconeogenesis, interconversion of pentoses and hexoses. Lipids :Classification, structure and functions, biosynthesis of fatty acids, oxidation of lipids, triglycerides, phospholipids, sterols.

#### Unit-3:

Biophysical Techniques:Principles and applications of thin layer chromatography, gas chromatography, HPLC and FPLC.Principles and applications of X-ray diffraction, NMR, ESR, ORD/CD, fluorescence, UV, IR, visible, mass spectroscopy.

#### Unit-4:

Principles and applications of tracer techniques in biology: Effect of radiation on biological systems, radioactive isotopes and their half-life, autoradiography, Radiation dosimetry, Cerenkov radiation, liquid scintillation spectrometry.

#### Books Recommended:

- ❖ Biochemistry by Stryer, Freeman publisher
- ❖ Biochemistry, Vol I, II, III by Geoffrey Zubey, WCB press
- ❖ Fundamentals of Biochemistry by Voet, Voet & Pratt, John Wiley publisher
- ❖ Principles of biochemistry by Albert Lehninger, David L Nelson & Michael M Cox, Mac Milan worth publisher.



# CELL AND MOLECULAR BIOLOGY

## (MBT-C-103)

### THEORY

#### Unit-1:

Structural and molecular organization of prokaryotic and eukaryotic cells; Functions of organelles; Cytoskeleton and cell motility; Cell cycle and division; Programmed cell death. Maternal gene effects; Embryogenesis; Cell lineages; Stem cell differentiation; Blood cell formation; Fibroblasts and their differentiation; Differentiation of cancerous cells.

#### Unit-2:

Concepts of replication in prokaryotes and eukaryotes; Enzymes involved in replication; Replication of single standard circular DNA; DNA repair; Recombination; homologous and non-homologous recombination; Gene targeting; Gene disruption.

#### Unit-3:

Prokaryotic and Eukaryotic transcription and regulation: Promoters, Regulatory elements, Operons, Processing of tRNA and rRNA; Translation machinery; Universal genetic code, Degeneracy of codons; Stop codons; Wobble hypothesis; Post translational modifications; Protein synthesis, transport and stability.

#### Unit-4:

Nonsense, missense and point mutations; Frameshift mutations; Mutagens; Transposons in prokaryotes and eukaryotes; Viral and cellular oncogenes; Tumor suppressor genes from humans; Activation of oncogenes; Suppression of tumor suppressor genes; Oncogenes as transcriptional activators.

#### Books Recommended:

- ❖ Essential Cell Biology by *Bruce Alberts et al.*, Garland Publisher.
- ❖ Cell and Molecular Biology by *F. D P deRobertes*, LW & W Publisher.
- ❖ Molecular Biology of the Cell by *Alberts, Bray, Lewis, Raff, Roberts and Watson*, Garland Publishers.
- ❖ Molecular Cell Biology by *H. Lodish, D. Baltimore, A. Bark, S. L. Zipursky, P. Matsudaira and J. Darnell*, Scientific American Books.



# GENOMICS AND PROTEOMICS (MBT-E-101) THEORY

## Unit-1:

*Introduction:* Structural organization of genome in Prokaryotes and Eukaryotes; Organelle DNA-mitochondrial, chloroplast; DNA sequencing-principles and translation to large scale projects; Recognition of coding and non-coding sequences and gene annotation; Tools for genome analysis-RFLP, DNA fingerprinting, RAPD, PCR, Linkage and Pedigree analysis-physical and genetic mapping.

## Unit-2:

*Genome sequencing projects:* Microbes, plants and animals; Accessing and retrieving genome project information from web; Comparative genomics, Identification and classification using molecular markers-16S rRNA typing/sequencing, ESTs and SNPs.

## Unit-3:

*Proteomics:* Protein analysis (includes measurement of concentration, amino-acid composition, N-terminal sequencing); 2-D electrophoresis of proteins; Microscale solution isoelectric focusing; Peptide fingerprinting; LC/MS-MS for identification of proteins and modified proteins; MALDI-TOF; SAGE and Differential display proteomics, Protein-protein interactions, Yeast two hybrid system.

## Unit-4:

*Functional genomics and proteomics:* Analysis of microarray data; Protein and peptide microarray-based technology; PCR-directed protein *in situ* arrays; Structural proteomics

## Books recommended;

- ❖ Genomes by *T.A. Brown*, John Wiley & Sons Ltd, New York
- ❖ Genome analysis (Volume I, II, III and IV) a Laboratory Manual by *Bruce Birren, Eric D. Green, Sue Klapholz, Richard M. Myers and Jane Roskams*, Cold Spring Harbor Laboratory Press.
- ❖ Discovery Genomics, Proteomics and Bioinformatics, *Campbell AM & Heyer L*, 2004, Pearson Education.
- ❖ Discovery genomics, proteomics and bioinformatics, *Campbell AM & Heyer L*, 2004, Pearson education.
- ❖ Methods in proteome and protein analysis, *Kamp RM*, 2004, Springer.



# **BIOENTREPRENEURSHIP**

## **(MBT-E-102)**

### **Unit-1:**

Meaning, Needs and Importance of Entrepreneurship, Promotion of entrepreneurship, Factors influencing entrepreneurship, Features of a successful entrepreneurship.

### **Unit-2:**

Forms of Business Organization, Project Identification, Selection of the product, Project formulation, Assessment of project feasibility.

### **Unit-3:**

Importance of Finance / Loans and repayments, Characteristics of Business finance, Fixed capital management: Sources of fixed capital, working capital its sources and how to move for loans, Inventory direct and indirect raw materials and its management.

### **Unit-4:**

Meaning and Importance, Marketing-mix, product management – Product line, Product mix, stages of product like cycle, marketing Research and Importance of survey, Physical Distribution and Stock Management. Meaning of International business, Selection of a product, Selection of a market for international business, Export financing, Institutional support for exports.

### **Books Recommended:**

- Holt DH. Entrepreneurship: New Venture Creation.
- Kaplan JM Patterns of Entrepreneurship.
- Gupta CB, Khanka SS. Entrepreneurship and Small Business Management, Sultan Chand & Sons.



## PRACTICAL BASED ON PAPER MBT-C-101, MBT-C-102&MBT-C-103

### (MBT-C-104)

1. Plasmid DNA isolation and DNA quantitation: Plasmid minipreps
2. Restriction digestion
3. Preparation of competent cells.
4. Agarose gel electrophoresis
5. Restriction Enzyme digestion of DNA
6. Purification of DNA from an agarose gel
7. DNA Ligation
8. Transformation of *E.coli* with standard plasmids, Calculation of transformation efficiency
9. Cloning of genomic DNA in standard plasmid vectors
10. Confirmation of the insert, Miniprep of recombinant plasmid DNA, Restriction mapping
11. Polymerase Chain reaction, using standard 16srRNA eubacterial primers
12. RFLP analysis of the PCR product
13. Transformation of yeast *Saccharomyces cerevisiae*
14. To prepare an Acetic-Na Acetate Buffer system and validate the Henderson-Hasselbach equation.
15. To determine an unknown protein concentration by plotting a standard graph of BSA using UV-Vis
16. Spectrophotometer and validating the Beer- Lambert's Law.
17. Titration of Amino Acids and separation of aliphatic, aromatic and polar amino acids by TLC.
18. Biophysical methods (Circular dichroism spectroscopy, fluorescence spectroscopy).
19. Determination of mass of small molecules and fragmentation patterns by Mass Spectrometry
20. Sterilization, disinfection, safety in microbiological laboratory.
21. Preparation of media for growth of various microorganisms.
22. Identification and culturing of various microorganisms.
23. Staining and enumeration of microorganisms.
24. Growth curve, measure of bacterial population by turbidometry and studying the effect of temperature, pH, carbon and nitrogen.
25. Assay of antibiotics production and demonstration of antibiotic resistance.
26. Isolation and screening of industrially important microorganisms.
27. Determination of thermal death point and thermal death time of microorganisms.
28. Microscopy – Bright field, dark field, phase contrast, fluorescence microscopy, visit to Electron microscope and Confocal microscope facilities.
29. Histology – Hand-sectioning of stem and leaf, saffranin and fast green staining.
30. Microtomy - fixing of tissues, dehydration, wax-embedding, sectioning and staining.
31. Mitosis – Onion root tips.
32. Meiosis – Insect testes





## M. Sc (First Year) SEMESTER-II

### RECOMBINANT DNA TECHNOLOGY

(MBT-C-201)

#### THEORY

##### Unit 1:

Enzymes used in recombinant DNA- technology Restriction endonucleases, isoschizomers, DNA methylases, DNA polymerases, DNA dependent RNA polymerases, ligases, kinases and phosphatases, nucleases, DNA binding proteins, single stranded – DNA binding proteins, DNA-repairing enzymes and topoisomerases.

##### Unit 2:

Vectors for molecular cloning Plasmids, bacteriophages, cosmids, single stranded filamentous bacteriophages, charon phages, shuttle vectors. Vectors used for cloning in bacteria, yeast, plants and animal cells. Strategies for cloning, Genomic and cDNA library preparation and screening strategies.

##### Unit 3:

Sequencing strategy DNA sequencing, Sanger's method, Maxim and Gilberts method, Next Generation Sequencing Technologies, Site directed mutagenesis and PCR technology: Principles, Variations and Applications. Microarray Technology: Fabrication of Chips, Applications

##### Unit 4:

Expression of cloned genes Expression vectors for mammalian cells, yeast cells, plant cells and bacterial cells (*E. coli*, *Bacillus*, *Streptomyces*), introduction of recombinant vectors into the expression hosts (mammalian cells, plant cells, yeast cells and bacterial cells,) cloned gene product expressed as fusion proteins expressed from cloned genes- production of antibodies, purification of antibodies, purification technology.

##### Books Recommended:

- ❖ Recombinant DNA, J.D. Watson, J. Witkowski & M. Gilman, M. Zoller, Scientific American Books New York.
- ❖ Principles of Gene Manipulation, W. Old and S. B. Primrose.
- ❖ Basic Methods in Molecular Biology, L.G. Davis, M.D. Dibner, J.R. Battey, Elsevier, New York, London.
- ❖ Molecular Cloning. A laboratory Manual, (Vol-I-III). J. Sambrook, E.F. Fritsch, T. Maniatis, Cold Spring Harbor Laboratory PRESS.
- ❖ Molecular Biology of Gene, J.D. Watson; J. NN.Roberts, N.H. Hopkins J.A. Steitz; A.N. Weiner, Benjamin/Cummings Pub. Cop. Inc. USA.



# IMMUNOLOGY AND IMMUNOTECHNOLOGY (MBT-C-202)

## Unit-1:

Components of innate and acquired immunity; Phagocytosis; Complement and Inflammatory responses; Haematopoiesis; Organs and cells of the immune system- primary and secondary lymphoid organs; Lymphatic system; Lymphocyte circulation: Lymphocyte homing; Mucosal and Cutaneous associated Lymphoid tissue. (MALT & CALT); Mucosal Immunity; Antigens - immunogens, haptens; Major Histocompatibility Complex - MHC genes, MHC and immune responsiveness and disease susceptibility, HLA typing

## Unit-2:

Immunoglobulins-basic structure, classes & subclasses of immunoglobulins, antigenic determinants; Multigene organization of immunoglobulin genes; B-cell receptor; Immunoglobulin superfamily; Principles of cell signaling; Basis of self -non-self discrimination; Kinetics of immune response, memory; B cell maturation, activation and differentiation; Generation of antibody diversity; T-cell maturation, activation and differentiation and T-cell receptors; Functional T Cell Subsets; Cell-mediated immune responses, ADCC; Cytokines-properties, receptors and therapeutic uses; Antigen processing and presentation- endogenous antigens, exogenous antigens, non-peptide bacterial antigens and super-antigens; Cell-cell co-operation, Hapten-carrier system

## Unit-3:

Precipitation, agglutination and complement mediated immune reactions; Advanced immunological techniques- RIA, ELISA, Western blotting, ELISPOT assay, immunofluorescence, flow cytometry and immunoelectron microscopy; Surface plasmon resonance, Biosensor assays for assessing ligand -receptor interaction, CMI techniques- lymphoproliferation assay, Mixed lymphocyte reaction, Cell Cytotoxicity assays, Apoptosis, Microarrays, Transgenic mice, Gene knock outs

## Unit-4:

Active and passive immunization; Live, killed, attenuated, sub unit vaccines; Vaccine technology- Role and properties of adjuvants, recombinant DNA and protein based vaccines, plant-based vaccines, reverse vaccinology; Peptide vaccines, conjugate vaccines; Antibody genes and antibody engineering- chimeric and hybrid monoclonal antibodies; Catalytic antibodies and generation of immunoglobulin gene libraries.

## Books Recommended:

- ❖ Cellular and Molecular Immunology by *Abbas et. al.*, Saunder Publication.
- ❖ Essential Immunology by *Roitt*, Blackwell Publisher.
- ❖ Immunology by *Kuby*, Freeman Publisher.
- ❖ Immunology-a short course by *Benjamini*, Wiley-Liss Publisher.

# BIOINFORMATICS AND BIostatISTICS

## (MBT-C-203)

### Unit-1:

Fundamental concepts in applied probability; Exploratory data analysis and statistical inference; Probability and analysis of one and two way samples; discrete and continuous probability models; Expectation and variance; Central limit theorem; Inference; Hypothesis; Critical region and error probabilities; Tests for proportion; Equality of proportions; equality of means of normal populations (variance known, variance unknown); Chi-square test for independence; P-value of the statistic; Confidence limits; Introduction to one way and two-way analysis of variance; Data transformations

### Unit-2:

Elements of programming languages - C and PERL; Data base concept; Database management system; Database browsing and Data retrieval; Sequence database and genome database; Data Structures and Databases; Databases such as GenBank; EMBL; DDBJ; Swissprot; PIR; MIPS; TIGR; Hovergen; TAIR; PlasmoDB; ECDC; Searching for sequence database like FASTA and BLAST algorithm.

### Unit -3:

Cluster analysis; Phylogenetic clustering by simple matching coefficients; Sequence Comparison; Sequence pattern; Regular expression based pattern; Theory of profiles and their use in sequence analysis; Markov models; Concept of HMMs; Baum-Welch algorithm; Use of profile HMM for protein family classification; Pattern recognition methods

### Unit-4:

Methods for modeling; Homology modeling; Threading and protein structure prediction; Structure-structure comparison of macromolecules with reference to proteins; Force fields; Molecular energy minimization; Monte Carlo and molecular dynamics simulation

### Books Recommended:

- ❖ Wayne W. Daniel, Biostatistics : A foundation for Analysis in the Health Sciences, 8th Edition, Wiley, 2004.
- ❖ Prem S. Mann, Introductory Statistics, 6th Edition, Wiley, 2006.
- ❖ John A. Rice, Mathematical Statistics and Data Analysis, 3rd Edition, John A. Rice, Duxbury Press, 2006.
- ❖ Campbell and Heyer, Discovering Genomics, Proteomics, & Bioinformatics, 2nd Edition, Benjamin Cummings, 2002.
- ❖ Introduction to Bioinformatics by *Stephen A Krawetz and David D. Womble*, Humana Press.
- ❖ Bioinformatics: Sequence and Genome Analysis by *David W. Mount*, Cold Spring Harbor Laboratory Press
- ❖ Fundamental of Biostatistics (5<sup>th</sup> edition) by *Bernard Rosner*, Duxbury Thomson Learning.
- ❖ Basic Statistics (2<sup>nd</sup> edition) by *B. L. Agrawal*, Wiley Eastern India.
- ❖ Introductory Statistics for Biology Students by *T. A Hall*, Chapman & Hall publisher.
- ❖ Statistical Methods in Biology by *N. T. J Bailey*, Cambridge Press.



# IPR AND BIOSAFETY ISSUES (MBT-E-201)

## Unit-1:

Types of IP: Patents, Trademarks, Copyright & Related Rights, Industrial Design, Traditional Knowledge, Geographical Indications, Protection of New GMOs; International framework for the protection of IP, IP as a factor in R&D; IPs of relevance to Biotechnology and few Case Studies; Introduction to History of GATT, WTO, WIPO and TRIPS

## Unit-2:

Invention in context of "prior art"; Patent databases; Searching International Databases; Country-wise patent searches (USPTO, EPO, India etc.); Analysis and report formation, Types of patents; Indian Patent Act 1970; Recent Amendments; Filing of a patent application; Precautions before patenting-disclosure/non-disclosure; WIPO Treaties; Budapest Treaty; PCT and Implications; Role of a Country Patent Office; Procedure for filing a PCT application

## Unit-3:

Patent application- forms and guidelines, fee structure, time frames; Types of patent applications: provisional and complete specifications; PCT and convention patent applications; International patenting-requirement, procedures and costs; Financial assistance for patenting-introduction to existing schemes; Publication of patents-gazette of India, status in Europe and US Patenting by research students, lecturers and scientists-University/organizational rules in India and abroad, credit sharing by workers, financial incentives Patent infringement- meaning, scope, litigation, case studies and examples

## Unit-4:

Introduction; Historical Background: Introduction to Biological Safety Cabinets; Primary Containment for Biohazards; Biosafety Levels; Biosafety Levels of Specific Microorganisms; Recommended Biosafety Levels for Infectious Agents and Infected Animals; Biosafety guidelines - Government of India; Definition of GMOs & LMOs; Roles of Institutional Biosafety Committee, RCGM, GEAC etc. for GMO applications in food and agriculture; Environmental release of GMOs; Risk Analysis; Risk Assessment; Risk management and communication; Overview of National Regulations and relevant International Agreements including Cartagena Protocol.

### *Important Links*

<http://www.w3.org/IPR/>

<http://www.wipo.int/portal/index.html.en>

[http://www.ipr.co.uk/IP\\_conventions/patent\\_cooperation\\_treaty.html](http://www.ipr.co.uk/IP_conventions/patent_cooperation_treaty.html) [www.patentoffice.nic.in](http://www.patentoffice.nic.in)

[www.iprlawindia.org/](http://www.iprlawindia.org/) - 31k - Cached - Similar page

<http://www.cbd.int/biosafety/background.shtml>

<http://www.cdc.gov/OD/ohs/symp5/jyrtext.htm>

<http://web.princeton.edu/sites/ehs/biosafety/biosafetypage/section3.html>



## MOLECULAR BREEDING (MBT-E-202)

### Unit-1:

Plant Genome – Nuclear and cytoplasmic; Significance of organelle genomes; Genome size and sequence components; Modern gene concept - Gene structure, structural and functional genes. Molecular markers – Restriction based and PCR based; DNA profiling using different assays- RFLP, RAPD, AFLP, ISSR, SNP etc. Development of SCAR and SSR markers.

### Unit-2:

Gene flow in plants – Development of mapping population - Marker Assisted Selection (MAS), screening and validation; Trait related markers and characterization of genes involved; Mapping genes on specific chromosomes; QTL mapping; Gene pyramiding; Transcript mapping techniques. Development of ESTs

### Unit-3:

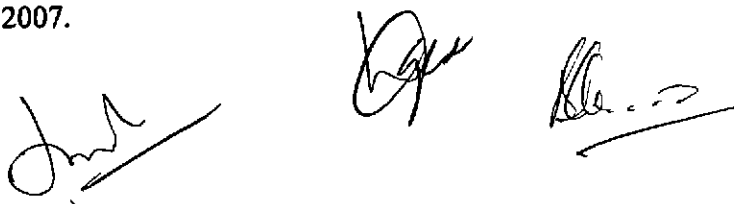
Molecular markers for plant genotyping and germplasm analysis; Fidelity analysis; settling IPR issues; Marker Assisted Breeding in transgenics – herbicide resistance; Pest and disease resistance; Quality enhancement etc.

### Unit-4:

Recent advances – Non gel based techniques for plant genotyping – Homogenous assays – Qualitative/Real Time assays; DNA Chip and its technology.

### Books Recommended:

- ❖ Anolles, G. C. and Gresshoff, P.M., DNA markers – protocols, applications and overviews. Wiley – Liss, New York, 1997
- ❖ Clark, D. P., Molecular Biology, Elsevier, USA, 2005.
- ❖ Henry R. J., Plant Genotyping: The DNA fingerprinting of plants. CABI, New Delhi, 2005.
- ❖ Patterson, Molecular dissection of complex traits, CRC Publications, Washington, 1998.
- ❖ Purohit, S. S., Biotechnology – Fundamentals and Applications, 8th Edition, Agrobios, India, 2007.



## PRACTICAL BASED ON PAPER MBT-C-201, MBT-C-202&MBT-C-203

### (MBT-C-204)

1. Selection of animals, Preparation of antigens. Immunization and methods of bleeding. Serum separation, Storage.
2. Antibody titre by ELISA method.
3. Double diffusion, Immuno-electrophoresis and Radial Immuno diffusion.
4. Complement fixation test.
5. Isolation and purification of IgG from serum or IgY from chicken egg.
6. SDS-PAGE, Immunoblotting, Dot blot assays
7. Blood smear identification of leucocytes by Giemsa stain
8. Separation of leucocytes by dextran method
9. Demonstration of Phagocytosis of latex beads
10. Separation of mononuclear cells by Ficoll-Hypaque
11. Flowcytometry, identification of T cells and their subsets
12. Lymphoproliferation by mitogen / antigen induced
13. Lymphnode Immunohistochemistry (direct and indirect peroxidase assay)
14. Hybridoma technology and monoclonal antibody production.
15. Immunodiagnosics using commercial kits
16. Isolation of nucleic acid from bacteria/viruses/eukaryotic cells
17. Amplification of the desired gene/fragment of the gene.
18. Analysis of the PCR/reverse transcriptase-PCR product in agarose gel.
19. Preparation of cloning vector (plasmid) from *E. coli* DH5 $\alpha$  and gel analysis.
20. Restriction digestion of the plasmid with suitable restriction endonuclease.
21. Insertion of the desired gene/gene segment and ligation with the help of ligase
22. Transformation of *E. coli* DH5 $\alpha$
23. Isolation of plasmid and confirmation of the insert by touch PCR and RE digestion
24. Cloning of gene in expression vector and Transformation.
25. Expression of protein in suitable host system.
26. Analysis of recombinant protein in SDS-PAGE
27. Purification of protein on Ni-NTA column and analysis of purified protein in SDS-PAGE
28. Sequence information resource
29. Understanding and use of various web resources: EMBL, Genbank, Entrez, Unigene, Protein information resource (PIR)
30. Understanding and using: PDB, Swissprot, TREMBL
31. Using various BLAST and interpretation of results.
32. Retrieval of information from nucleotide databases.
33. Sequence alignment using BLAST.
34. Multiple sequence alignment using Clustal W.



## **M. Sc (Final Year) SEMESTER-III**

### **FERMENTATION TECHNOLOGY AND DOWNSTREAM PROCESSING (MBT-C-301) THEORY**

#### **Unit-1:**

Fermentation: Solid State (SSF) and Submerged (SmF) fermentations, Media design and optimization, Basic requirements of fermentative microorganisms, carbon and nitrogen sources for large-scale fermentation, optimization of media and product information. Reactor design and instrumentation Batch, continuous and plug flow reactor systems, conventional stirred tank reactor (CSIR), bubble column reactor, air lift reactor, packed bed reactor, fluidized bed reactor, tube plug reactor, sterilization of bioreactors, aseptic function of bioreactor and instrumentation, bioreactors for plant, algal and animal cell cultures.

#### **Unit-2:**

Kinetic models and mass transport in bioreactors A general mass balance of the reactor, biological reaction rate equation, maintenance and cell yield, determination of parameters of rate equation, oxygen demand and transport, controlled oxygen transfer rate and analysis, internal diffusion in solid phase catalysis external diffusion at solid phase liquid interphases.

#### **Unit-3:**

Modes of process operation and scale up Batch, continuous & fed-batch fermentation, enzymatic processes, rheology of fermentation broth, power calculations, calculations of KLa, scale up criteria. Down stream processing Centrifugation, filtration, cell disintegration, extraction in aqueous two phase system, liquid – liquid extraction, fractional precipitation, fractional distillation, chromatographic and electrophoretic techniques.

#### **Unit-4:**

Integrated process analysis of some bioprocess technology products Salient features of beer brewing, fermented milk products, fermented Indian foods and vegetables, wine production, production of baker's yeast, ethanol production, microbial transformations, production of citric acid, antibiotics, polysaccharides, enzymes, vitamins, glutamic acid, single cell protein (SCP) and recombinant proteins.

#### **Books Recommended:**

- ❖ Bioprocess Technology: Fundamentals and Application, S.O. Enfors and L. Haggstrom, Royal Institute of Technology, Stockholm.
- ❖ Biotechnology: A Text Book of Industrial Microbiology, T. D. Brock; W. Grueger and A. Grueger: Sinauer Publication, USA.
- ❖ Biochemical Engineering, J.M. Lee, Prentice Hall International Ltd, U. K. London.
- ❖ Principles of Fermentation Technology, P. F. Stanburry and A. Whitaker.
- ❖ Manual of Industrial Microbiology and Biotechnology, A. L. Demain and N. A. Solomon, American Society for Microbiology, Washington



# ENVIRONMENTAL BIOTECHNOLOGY

## (MBT-C-302)

### THEORY

#### Unit 1:

Concept of Environmental Pollution; Origin of pollution; Classification and nature of Environmental Pollutants; Major sources; Impacts of Environmental Pollution at local regional and global level. Concept of air Pollution; Major air pollutants and their sources; Effects of air pollution on human health, animals, plants, material and climate; Formation of fog and photochemical smog and acid rain; Monitoring of air pollution: Control on release of smoke; Gaseous contaminants and odour; Control on release of particulate matter by using different control devices. Noise, Radiation and Soil Pollution: Causes and control measures.

#### Unit 2:

Waste water Treatment: Biological treatment system (Oxidative ponds, aerobic and anaerobic ponds, facultative ponds, aerated ponds), Biological waste treatment, activated sludge treatment, microbial pollution in activated sludge, percolating filters, waste water treatment by biofilms. Treatment scheme of Dairy, Distillery, Tannery, Sugar, Fertilizers, Refinery, Chemical and Antibiotic waste.

#### Unit 3:

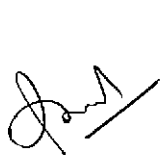
Bioremediation: *In situ* and *Ex situ* bioremediation; Constrains and priorities of bioremediation; Evaluating Bioremediation; Bioremediation of VOCs. Biodegradation; Factors affecting process of biodegradation; Methods in determining biodegradability; Contaminant availability for biodegradation. Xenobiotics; Persistence and biomagnification of xenobiotic molecules; Microbial interactions with xenobiotics; Phase I and Phase II reactions; Cyt P 450 mediated reactions; Use of microbes (bacteria and fungi) and plants in biodegradation and Biotransformation.

#### Unit 4:

Concept of solid waste; Industrial solid waste; Domestic solid waste; Agricultural solid waste; Municipal solid waste; Major sources of solid wastes; Effects of solid waste generation on quality of air, water and public health; Technical approach for solid waste management; Disposal of organic and medical waste; Recovery and recycling of metallic waste; Disposal of plastic waste and hazardous wastes. Sources of heavy metal pollution; Microbial interactions with inorganic pollutants - Microbial metal resistance; Microbial transformation; Accumulation and concentration of metals; Biosorption - Biotechnology and heavy metal pollution; Oil field microbiology; Improved oil recovery; Biotechnology and oil spills; Hydrocarbon degradation

#### Books recommended;

- ❖ Biotechnology – Expanding Horizons by *B.D. Singh*. 2<sup>nd</sup> Edition Kalyani Publishers.
- ❖ Microbial Ecology: Fundamentals & Applications by *Atlas, R.M.* Wc Brown.
- ❖ Environmental Microbiology by *A.H. Varman*, ASM Press.
- ❖ Biodegradation and Bioremediation by *Alexandar, M.* Wiley International.





# ENZYMOLGY AND ENZYME TECHNOLOGY (MBT-C-303) THEORY

## Unit-1:

Commercial sources of enzymes: Sources of commercial enzymes, microbial enzymes, screening strategies for isolation of hyper-enzyme producing microbes, control of microbial enzyme production, genetic manipulation techniques. Extraction and purification of enzymes Enzyme extraction, enzyme purifications, large-scale purification enzyme specification, criteria of purity, molecular weight determination and characterization of enzyme.

## Unit-2:

Enzyme kinetics: Nomenclature of enzymes, simple and complex (bisubstrate) enzyme, inhibition of enzyme reactions, factors effecting enzyme activity, enzyme reactors with simple kinetics.

## Unit-3:

Immobilization of enzymes Immobilization techniques, effect of mass transfer resistance, kinetics of immobilized enzymes, immobilization of amylase, cellulase, protease and lipase. V. Biocatalyst stabilization: Immobilization, medium engineering, enzyme reactions in water restricted media and super critical fluids. Use of additives chemical modifications and protein engineering for enzyme stabilization.

## Unit-4:

Enzyme engineering: Design and specialized construction of novel enzymes, synthetic enzymes, covalent modifications of enzymes, enzymic modifications of enzymes, substitution of bound metals in enzymes, non-and site-specific mutagenesis for the construction of desired enzymes. Specialized biocatalysts: Abzymes and Ribozymes.

## Books Recommended:

- ❖ Biocatalysts for industry, J.S. Dordick, Plenum Press, New York, London.
- ❖ Enzyme Technology, M.F. Chaplin & C. Bucke.
- ❖ Enzymes in Industry, Production & Applications W. Gerhartz.
- ❖ Enzymes Technology, P. Gacesa and J. Hubble, Open University Press, England, UK.
- ❖ Biochemical Engineering, J.M. Lee, Prentice Hall INC, New Jersey



# PLANT TISSUE CULTURE AND TRANSGENIC TECHNOLOGY (MBT-E-301) THEORY

## Unit-1

Totipotency; Tissue culture media; Plant hormones and morphogenesis; Direct and indirect organogenesis; Direct and indirect embryogenesis; Cell suspension culture; Micropropagation – shoot tip culture, somatic embryos, artificial seeds; Applications of tissue culture; Virus elimination by shoot tip culture; hybridization and embryo culture; Anther culture and dihaploids.

## Unit-2:

Large-scale cell suspension culture; Production of alkaloids and other secondary metabolites; Protoplast culture, Protoplast isolation and purification; Protoplast viability test; Protoplast culture; Protoplast fusion; Somatic hybrids; Cybrids. Direct transformation of protoplasts using PEG; electroporation; Transformation by particle bombardment; Assembly of particle gun; Microprojectile preparation and bombardment; Chloroplast transformation by particle bombardment

## Unit-3:

*Agrobacterium* biology; Ti plasmid-based transformation; crown gall and hairy root disease, Ti and Ri plasmids, T-DNA genes, borders, overdrive, chromosomal and Ti plasmid virulence genes and their functions, *vir* gene induction, mechanism of T-DNA transfer; Ti plasmid vectors, *vir* helper plasmid, super virulence and monocot transformation, binary vector; Floral dip transformation; Promoters and polyA signals; Protein targeting signals; Plant selectable markers; Reporter genes; Positive selection; Selectable marker elimination; Transgene silencing; Strategies to avoid transgene silencing.

## Unit-4:

Genetic engineering of crops; Commercial status of transgenic plants; Herbicide resistance, glyphosate, sulfonyl urea, phosphinothricin, atrazine; Pest resistance, *Bt* toxin, synthetic *Bt* toxin; Protease inhibitor; GNA and other lectins;  $\alpha$ -amylase inhibitor; nematode resistance; Genetic engineering for male sterility- Barnase-Barstar; Delay of fruit ripening; polygalacturanase, ACC synthase, ACC oxidase; Improved seed storage proteins; Improving and altering the composition of starch and plant oils; Golden rice for  $\beta$ - carotene accumulation; Production of antibodies and pharmaceuticals in plants; Bio-safety concerns of transgenic plants.

## Books Recommended:

- ❖ Plant Tissue Culture: Application and Limitation by *S. S. Bhojwani and M. K. Razdan*, Elsevier Publication
- ❖ Plants, Genes and Agriculture by *Maarten J Chrispeels and David E. Sadava*, Jones & Bartlett Publishers
- ❖ An Introduction to Plant Tissue Culture by *M. K. Razdan*, Oxford & IBH Publishing Co. Pvt. Ltd.
- ❖ Plant Biotechnology: The genetic manipulation of plants by *Adrian Slater, Nigel Scott, and Mark Fowler*, Oxford University Press



# ANIMAL CELL CULTURE TECHNOLOGY (MBT-E-302) THEORY

## Unit-1:

Cell culture Laboratory design & Equipments. Washing, packing and sterilization of different materials used in animal cell culture; Aseptic concepts; Maintenance of sterility; Cell culture vessels. Media and reagents Types of cell culture media; Ingredients of media; Physiochemical properties; CO<sub>2</sub> and bicarbonates; Buffering; Oxygen; Osmolarity; Temperature; Surface tension and foaming; Balance salt solutions; Antibiotics, growth supplements; Foetal bovine serum; Serum free media; Trypsin solution; Selection of medium and serum; Conditioned media; Other cell culture reagents; Preparation and sterilization of cell culture media, serum and other reagents.

## Unit-2:

Different types of cell cultures, History of animal cell culture; Different tissue culture techniques; Types of primary culture; Chicken embryo fibroblast culture; Chicken liver and kidney culture; Secondary culture; Trypsinization; Cell separation; Continuous cell lines; Suspension culture; Organ culture etc.; Behavior of cells in culture conditions: division, growth pattern, metabolism of estimation of cell number; Development of cell lines; Characterization and maintenance of cell lines, stem cells; Cryopreservation; Common cell culture contaminants.

## Unit-3:

Applications Cell cloning and selection; Transfection and transformation of cells; Commercial scale production of animal cells, stem cells and their application; Application of animal cell culture for in vitro testing of drugs; Testing of toxicity of environmental pollutants in cell culture; Application of cell culture technology in production of human and animal viral vaccines and pharmaceutical proteins.

## Unit-4:

Scale-up Cell culture reactors; Scale-up in suspension; Scale and complexity; Mixing and aeration; Rotating chambers; Perfused suspension cultures; Fluidized bed reactors for suspension culture; Scale-up in monolayers; Multisurface propagators; Multiarray disks, spirals and tubes; Roller culture; Microcarriers; Perfused monolayer cultures; Membrane perfusion; Hollow fiber perfusion; Matrix perfusion; Microencapsulation; Growth monitoring

## Books recommended:

- ❖ Animal Cell Culture: A practical approach by R.I. Freshney, IRL press.
- ❖ Culture of animal cells: A manual of basic techniques by R.I. Freshney, Willy-Liss and So publication.
- ❖ Animal cell culture technique by Martin Clynes, Springer publication.



## **PRACTICAL BASED ON PAPER MBT-C-301, MBT-C-302&MBT-C-303**

### **(MBT-C-304)**

1. Determination of oxygen transfer rate and volumetric oxygen mass transfer coefficient (KLa) under variety of operating conditions in shake flask and bioreactor.
2. Determination of mixing time and fluid flow behaviour in bioreactor under variety of operating conditions.
3. Rheology of microbial cultures and biopolymers and determination of various rheological constants.
4. Production of microbial products in bioreactors.
5. Studying the kinetics of enzymatic reaction by microorganisms.
6. Production and purification of various enzymes from microbes.
7. Comparative studies of Ethanol production using different substrates.
8. Microbial production and downstream processing of an enzyme, e.g. amylase.
9. Various immobilization techniques of cells/enzymes, use of alginate for cell immobilization.
10. Production of microbial enzyme by SSF /SmF method
11. Preparation of cell-free lysates
12. Purification of enzyme: Ammonium Sulfate precipitation, Ion-exchange Chromatography, Gel Filtration, Affinity Chromatography, Generating a Purification Table, Assessing purity by SDS-PAGE Gel Electrophoresis, Assessing purity by 2-D gel Electrophoresis
13. Enzyme Kinetics parameters Km, Vmax and Kcat etc..
14. Estimation of halides in water samples by potentiometry.
15. Estimation of  $\text{Co}^{2+}$  and  $\text{Ni}^{2+}$  by colorimetry/spectrophotometry.
16. Estimation of sulphates by turbidometry.
17. Estimation of heavy metals in various samples by AAS.
18. Field visit to river/lake and waste water treatment plants.
19. Sampling techniques: wastewater analysis for physico-chemical characteristics such as pH, conductivity, TDS, DO, BOD, COD,  $\text{CO}_2$ , alkalinity, nutrients, chlorides, hardness, settlability of solids.
20. Vermicomposting: collection, preparation and analysis of composted material for NPK, moisture holding and microbial load.
21. Microorganisms form polluted environment/Soil /Water resources /Air
22. Microbial degradation of textile dyes/pesticides/hydrocarbons and oils
23. Bioremediation
24. Pollutant removal using microorganisms from industrial effluent.
25. Removal of oil spills form soil
26. Effect of pesticides on soil microorganisms
27. Pollution control
28. Activated sludge process



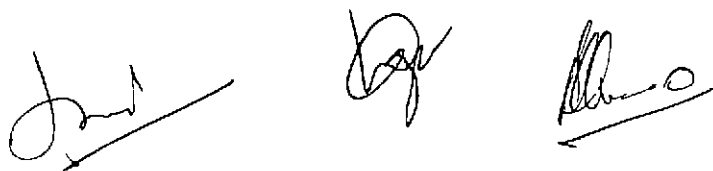
**M.Sc (Final Year) SEMESTER-IV**

**DISSERTATIONS  
(MBT-C-401)**

Each Student will have to submit an allotted **Dissertation** on specific topics related with different area of biotechnology and will submit a report on which *viva-voce* will be conducted.

**SEMINAR  
(MBT-C-402)**

Each student will have to present ONE Seminar on the topic ALLOTTED covering different areas of Biotechnology and will be evaluated by powerpoint presentation.

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