

‘School of Biotechnology’, Presidency University, Kolkata

Syllabus for M.Sc. in Biotechnology with specialization in A) Plant Biotechnology, and B) Microbial Biotechnology

Semester I*				
Course Code	Title	T/P	Credits	Marks
BT 0701*	Biochemistry, Enzymology, and Metabolism	T	4	50
BT 0702*	Molecular Biology, Cell Biology and Genetics	T	4	50
BT 0703*	Biostatistics and Bioinformatics	T	4	50
BT 0704*	Practical: Biochemistry, Enzymology, and Metabolism	P	4	50
BT 0705*	Practical: Molecular and Cell Biology	P	4	50

*These courses are common for both specializations of Biotechnology

SEMESTER I

BT 0701: Biochemistry, Enzymology, and Metabolism

Unit I: Carbohydrates and Lipids

pH and buffer, maintenance of blood pH, macromolecules, concepts of stereochemistry
Sugars- Monosaccharides and derivatives of sugars, polysaccharides, amylose, pectin and cellulose
Structure and properties of important members of storage and membrane lipids, glycerophospholipids
sphingolipids, cholesterol, lipid bilayer; Lipoproteins

Unit II: Structural properties of Nucleic Acids and Proteins

Structural elements of nucleic acids, specific properties of DNA and RNA, different types of RNA
structure
Primary, secondary, tertiary and quaternary structures of peptides and proteins, Ramachandran plot,
protein degradation, molecular pathways controlling protein degradation; Structural stability of proteins;
Protein folding, pathways of protein folding, chaperons.
Separation and analysis of nucleic acids and proteins, principles of protein purification

Unit III: Enzymes and enzyme technology

Structure and classification of enzymes, apoenzyme and cofactors, prosthetic group, coenzyme and metal
cofactors; Active site, activation energy, enzyme unit, specific activity, turnover number
Lock and key hypothesis, induced fit model, Enzyme kinetics and inhibition; Effect of temperature and
pH on enzyme activity.

Ribozymes, Abzymes, and Isozymes. Overview of protein-ligand interaction and their analyses.

Enzyme regulation- allosteric enzymes, allosteric modulators, feedback inhibition, kinetic properties of allosteric enzyme, Hill and Scatchard plots, regulation by covalent modification, regulation by proteolytic cleavage of protein, zymogens

Multienzyme systems and their regulation, principles of enzyme-enzyme interaction

Unit IV: Bioenergetics

Transport across membrane, ABC transporter, phosphotransferase system, drug export systems, amino acid transport.

Photosynthesis in bacteria and plants, photopigments, C3, C4 and CAM, metabolism of energy reserve compounds, metabolism of energy reserve compounds

Respiratory metabolism- Embden-Meyerhoff pathway, Entner–Doudoroff pathway, phosphoketolase pathway, glyoxylate pathway, Krebs cycle, oxidative and substrate level phosphorylation, reverse TCA cycle, gluconeogenesis- Pasteur effect.

Metabolic energetics: Electron transport system; basic mechanisms of ATP synthesis.

Auxotrophs, Anaerobic respiration and chemolithotrophy: nitrate, carbonate, sulfate as electron acceptors.

Biological N₂-fixation-ammonia assimilation, denitrification, molecular mechanism and genetic regulation of N₂-fixation; nitrogenase structure.

BT 0702: Molecular Biology, Cell Biology and Genetics

Unit I: Chromatin structure and modifications

Chromatin organization- histone and DNA interactome, structure and assembly of eukaryotic and prokaryotic DNA polymerases, DNA-replication, repair and recombination; chromatin control: gene transcription and silencing by chromatin- Writers, -Readers and -Erasers.

Unit II: RNA and Transcriptional control

Transcriptional control: structure and assembly of eukaryotic and prokaryotic RNA Polymerases, promoters and enhancers, transcription factors as activators and repressors, transcriptional initiation, elongation and termination.

Post-transcriptional control, selective and specific mRNAs through interference by small non-coding RNAs (miRNAs and siRNAs).

Unit III: Translational controls

Genetic code and its properties, Wobble hypothesis, prokaryotic and eukaryotic protein synthesis- initiation, elongation, termination; co- and post-translational modifications; Translation-dependent regulation of mRNA and protein stability

Unit IV: Cell biology

Cellular organization, organelles, cell wall, extracellular matrix; Cell-cell interaction; Cell-matrix interaction, Cell communication- Signaling molecules; Receptors: G-protein coupled receptor, Receptor Tyrosine Kinase, cytokine receptors; Pathways of intracellular signal transduction; Regulation of cellular activities (autophagy, degradative pathways). Programmed cell death, necrosis and senescence.

Nucleus and nuclear transport- import and export of protein, export of different RNAs

Cytoskeleton- microtubules, microfilaments, intermediate filaments, molecular motors, bacterial cytoskeleton

Cell cycle, and checkpoint regulation, differentiation; Genetic basis of cancers: Protooncogene, Oncogene, Tumor suppressor genes; Oncogenesis

Unit V: Microscopic techniques in Cell biology

Visualization of cells and subcellular components by light microscopy, resolving powers of different microscopes, microscopy of living cells, Electron microscopy, scanning and transmission electron microscopy, stereomicroscopy, image processing methods in microscopy.

Unit VI: Genetics

Model systems in genetic analysis (*E. coli*, *Neurospora crassa*, yeast, Arabidopsis, Rice, Drosophila) non-Mendelian and Mendelian ratios, Linkage mapping in haploids and diploids, Regulation of Gene expression: Operon concept, genetic switch in bacteriophage lambda.

Genetics of yeast, gene conversion, auxotrophic and temperature sensitive mutants

Quantitative Genetics; Population Genetics: introduction to the elements of population genetics: genetic variation, genetic drift, neutral evolution; mutation selection, balancing selection, Fishers theorem, Hardy-Weinberg equilibrium, linkage disequilibrium; in-breeding depression & mating systems; population bottlenecks, migrations, Bayesian statistics; adaptive landscape, spatial variation & genetic fitness.

BT 0703: Biostatistics and Bioinformatics

Unit I: Basics of Biostatistics

Basic definitions and applications of Biostatistics; Principles and practice of statistical methods in biological research; samples and populations; Data collection and representation- types of data, methods

of collection of primary and secondary data, methods of data representation, graphical representation by histogram, polygon, ogive curves and pie diagram. Measures of central tendency: mean, median, mode; Measures of dispersion- range, mean deviation, coefficient of variation; standard deviation, standard error.

Unit II: Application of Biostatistics

Experimental design, introduction to probability theory, probability distributions- binomial, poisson, and normal distribution; Statistical inference- hypothesis testing, significance level, test of significance for large (Z test) and small samples, correlation and regression; Positive and negative correlation and calculation of Karl-Pearson's coefficient of correlation. Partial correlation, calculation of an unknown variable using the regression equation. Spearman's rank correlation, nonparametric tests (Chi-square t test, G test), Mann-Whitney U test, ANOVA- one- and two-way; Use of biostatistics software.

Unit III: Introduction to Bioinformatics

Scope and applications of bioinformatics, global bioinformatics scenario, definition of terms- orthology, paralogy, xenology and analogy; Similarity and identity; Introduction to databases- types of databases, information retrieval system (Entrez and SRS) and database collaboration, file formats, sequence, structure and pathway databases of nucleotides and protein.

Unit IV: Computational Methods and Application of bioinformatics

Substitution scoring matrices - PAM, BLOSUM, Scoring Schemes, gap penalty, concept of distance and similarity matrix; Alignments - pairwise alignment, dynamic Programming, global alignment - Needleman and Wunsch, and local alignment - Smith-Waterman algorithms.

Multiple Sequence Alignment, progressive method, iterative method; data searching tools for homologous sequences analysis - BLAST & FASTA; Sequence editors - BioEdit, BoxShade; Prediction tools - profile, motifs, domains and feature identification; Phylogenetic prediction: Phylogenetic tree construction - distance based method and character-based methods; Gene prediction, protein structure & functions prediction, Phylogenetic analysis package - MEGA; Homology modeling: 2D and 3D protein modeling.

BT 0704: Practical: Biochemistry, Enzymology, and Metabolism

1. Determination of pH optima, K_m and V_{max} of enzyme alkaline phosphatase.
2. Estimation of DNA, RNA and Protein
3. Separation of proteins on denaturing SDS-PAGE and determination of molecular weight of an unknown protein
4. Extraction and analysis of Specific activity of peroxidase
5. Estimation of total alkaloids, total phenols, total flavonoids, and Phytosterols

6. Hands on experiments using protein structure, modeling, docking, drawing of phylogenetic tree

BT 0705: Practical: Molecular and Cell Biology

1. Plasmid DNA isolation, DNA quantitation, and Agarose gel electrophoresis
2. Concept of lac-operon: a) Lactose induction of β -galactosidase, b) Glucose Repression. c) Diauxic growth curve of *E. coli*
3. Preparation of competent *E. coli* cells; Transformation of *E. coli* with a known plasmid, and determination of transformation efficiency
4. Observation of bacteria, mammalian and plant cells/tissue section under light/compound microscope, determination of cell number by hemocytometer
5. Microscopic observation of cellular organelles

Syllabus for M.Sc. in Biotechnology with specialization in Plant Biotechnology (Semester II-IV)

Semester II				
BTPB 0801	Introduction to Plant system	T	4	50
BTPB 0802	Plant Genetic Engineering and Biotechnology			
BTPB 0803	Plant Stress Biology and Environmental Sciences	T	4	50
BTPB 0804	Practical: Plant Tissue Culture and Genetic Engineering	P	4	50
BTPB 0805	Practical: Plant Stress Biology	P	4	50
Semester III				
BTPB 0901	Plant Physiology and Plant Breeding	T	4	50
BTPB 0902	Plants Genomics for Crop Improvement	T	4	50
BTPB 0903	Transgenic Plants for Crop improvement	T	4	50
BTPB 0904	Practical: Plant Physiology and Plant Breeding	P	4	50
BTPB 0905	Practical: Plant Molecular Breeding and Transgenics	P	4	50
Semester IV				
BTPB 1001	Principles of Biomass and Bioenergy	T	4	50
BT 1002*	Biosafety and Bioethics, Intellectual Property Rights, and Bioentrepreneurship	T	4	50
BTPB 1003	Journal club and Group discussion	T	4	50
BTPB 1004	Lab Training and Dissertation on Plant biotechnology	P	4	50
BTPB 1005	Industrial Training	P	4	50

**This course is common for both specializations of Biotechnology*

SEMESTER II

BTPB 0801: Introduction to Plant system

Part A: Plant Tissue Culture

Unit I: Uniqueness of the Plant system

Photosynthesis, N₂ fixation, Vitamins, mineral sources, chloroplasts, mitochondria engulfing

Unit II: Prerequisite for Plant Tissue Culture

Sterilization procedures, fumigation, wet and dry sterilization, ultraviolet sterilization, ultra-filtration and surface sterilization design of laboratory and commercial tissue culture facility.

Unit III: Tissue Culture Media

Media for *in vitro* culture; types of media, solid, liquid and commercial prepacked media; media composition, macronutrients, micronutrients, and plant growth regulators; preparation of media; selection of suitable media.

Unit IV: Explants

Concept of totipotency; Callus culture; initiation and maintenance of callus, micropropagation, direct and indirect morphogenesis, somatic embryogenesis and synthetic seed production; haploid, doubled haploid and triploid culture; somaclonal variation induction and thin layer culture

Unit V: Suspension Culture

Culture systems, Isolation of single and aggregate of cells and regeneration of plants; Immobilization of cells and use of bioreactors and hairy root culture. Protoplast culture - Isolation of protoplast, culture of protoplast, regeneration and sub-protoplast; Somatic cell hybridization, selecting desired hybrids and their regeneration into plants.

Part B: Essential Techniques in Plant Biology

Unit I: Spectroscopy

Absorption, fluorescence and emission spectroscopy of biomolecules, UV Visible Spectroscopy, Fluorescence Spectroscopy.

Unit II: Centrifugation

Principle of centrifugation and different types of centrifuge. Differential & density gradient centrifugation

Unit III: Chromatography Techniques

TLC, HPLC, HPTLC & FPLC, Size-exclusion Chromatography, Affinity chromatography, Ion-exchange Chromatography

Unit IV: Structure Determination of Biomolecules

Structure determination of biomolecules using NMR spectroscopy and X-ray crystallography.

Unit V: Imaging Applications in Plants

Organelle-specific stains, intracellular localization of fluorescently-labeled chimeric proteins. Tissue preparation (Tissue fixation, Embedding and sectioning), staining and visualization; Localization of Fluorescent-tagged protein in the plant cell by transient expression analysis; Protein-interaction analysis by co-localization of fluorescent tagged proteins by FRET and or BiFC methods.

BTPB 0802: Plant Genetic Engineering and Biotechnology

Unit I: Basic tools in Genetic Engineering

Restriction enzyme, exonucleases, endonucleases, reverse transcriptase, polynucleotide kinase, DNA ligase, terminal deoxynucleotidyl transferase, alkaline phosphatase, ligation of DNA fragments with vectors homopolymer tailing, linkers and adaptors.

Unit II: Recombinant DNA Technology

Construction of genomic and cDNA libraries; Isolation of genes, chemical synthesis of genes and synthetic genomics. Vectors: cloning and expression vectors, types of vectors used in higher plants, Tumour-inducing (Ti) plasmids, binary and co-integrate vectors, Plant selection markers, reporter genes- GFP, luciferase, GUS

Unit III: Methods in Plant genetic engineering

DNA sequencing; Principles and methods, blotting techniques; Southern, Northern and Western. Polymerase chain reaction and methods, fusion PCR, degenerate and RACE PCR - Site-directed mutagenesis, transposon mutagenesis, fluorescence *in-situ* hybridization, electrophoretic mobility shift assay, DNA footprinting, restriction mapping, DNA fingerprinting and chromosome walking.

Unit IV: Introducing genes into prokaryotes

Target cell preparation, natural gene transfer methods, calcium chloride mediated transformation. Shotgun cloning, selection, and screening of recombinant clones, methods based on nucleic acid hybridization, finding specific clones by functional complementation, reporter genes and analysis of promoter for high expression of the transgene.

Unit V: Introducing genes into plants

Target cell preparation, prerequisites for transgenic plant production, *Agrobacterium*-mediated transformation, pollen-mediated gene transfer. Physical delivery methods: particle bombardment and microinjection, gene uptake to the protoplast. Chemical mediated DNA delivery; electroporation, liposome, and ultrasonication mediated methods, analysis of transgenics using Southern, Northern, Western blots, functional validation

BTPB 0803: Plant Stress Biology and Environmental Sciences

Unit I: Plant Stress Physiology

Types of environmental stresses; Plant responses to abiotic and biotic stresses.

Unit II: Abiotic Stress

Plant response to abiotic stress; drought and salt stress, osmotic adjustment and its role, acid soil stress, metal stress, waterlogging, light, cold and heat stress, stress-inducible proteins and genes. The role of plant growth regulators in stress tolerance mechanisms and nutrient deficiency stress and disorders in plants.

Unit III: Biotic Stress

Plant pathology; its scope and relationships to other sciences, concept of plant diseases, pathogenicity, pathogen penetration and entry, colonization in the host, factors affecting infection. Enzymes in plant diseases; cell wall degrading enzyme, toxins in relation to plant diseases, defense mechanism. Genetics of plant-pathogen interaction; effect of environment on diseases development, epidemiology, forms of epidemics and conditions governing some of the important crop diseases.

Unit IV: Plant Stress Molecular Biology

Stress sensors; signal transduction, MAPK pathway, CDPK, and other pathways. Transcriptional regulation of stress tolerance, MYB, WRKY, NAC, bZIP and other factors. Stress responsive gene expression and phenotypic responses; Hyper sensitive response (HR), systemic acquired response, ROS generation, programmed cell death.

Unit V: Pollution and Phytoremediation

Air pollution, water pollution (heavy metal pollution and thermal pollution) and soil pollution (pesticide pollution) (outline only). Phytoremediation; introduction, types of phytoremediation, bioremediation, factors influencing bioremediation and phytoremediation

BTPB 0804: Practical: Plant tissue culture and Genetic Engineering

1. Preparation of competent cells of *E. coli* / *Agrobacterium* spp. for transformation.
2. PCR based cloning of a DNA fragment, ligation into plasmid vector, transformation using ligation mix and selection of transformants.
3. Colony PCR assay to screen the positive *E. coli* / *Agrobacterium* spp. transformants containing the ligated product and restriction digestion of the positive clone.
4. Sequence (DNA/Protein) alignment-based database search, Multiple Sequence Alignment and Phylogenetic analysis and NCBI database introduction.
5. Transformation of tobacco plants by following leaf disc method.
6. RNA isolation, cDNA synthesis and amplification of a gene by PCR.

BTPB 0805: Practical: Plant Stress Biology

1. Proteins isolation and estimation from the given plant material
2. Electrophoresis of plant proteins under denaturing conditions followed by staining with Coomassie Brilliant Blue R-250 and Silver staining methods.
3. Proteins isolation for resolving isozymes using native, non-denaturing polyacrylamide gel electrophoresis.
4. Estimation of chlorophyll from the control and stressed plants.
5. Estimation of antioxidant enzymes /ROS from the control and stressed plants.
6. Analysis of plant root systems in control and stressed plants by using ImageJ software.
7. Analysis of root microbial flora from healthy and diseased (fungal or bacterial) plants.

SEMESTER III**BTPB 0901: Plant Physiology and Plant Breeding****Part A - Plant Physiology [25 marks]****Unit I: Water Uptake**

Imbibition, diffusion, osmosis, water potential and its components, translocation of water, ascent of sap, transpiration, stomatal physiology, guttation, water stress and its significance and translocation in phloem.

Unit II: Growth

Measurement of growth, growth curve, PGR, auxins, gibberellins, cytokinins and ethylene, role of hormones in fruit ripening, application of hormones in agriculture. Nitrogen fixation in plants, nutrient uptake mechanisms, transport and function, nutrient deficiencies, regulation of plant growth and yield by nutritional status, plant growth promoting rhizosphere

Unit III: Photomorphogenesis

Photoperiodism, vernalisation, photoreceptors, physiology of germination, types and methods to overcome dormancy.

Unit IV: Photosynthesis and Respiration

Photosynthetic pigment systems, radiant energy, cyclic and noncyclic electron transport, C₃, C₄ and CAM pathways, factors affecting photosynthesis, photorespiration. Respiration; aerobic and anaerobic, glycolysis, Krebs's cycle, oxidation - reduction potential, ATP synthesis, factors affecting respiration.

Part B- Plant Breeding [25 marks]**Unit I: Basic Concepts of Genetics**

Mendel's laws; monohybrid crosses and its modification (incomplete dominance, codominance, heterodominance, lethal genes and pleiotropism) and dihybrid cross. Laws of segregation in plant crosses, inbreeding, selfing, heterosis, maintenance of genetic purity, gene pyramiding.

Unit II: Gene Interaction and Sex-Linked Inheritance

Complementary genes, supplementary genes, epistatic genes, duplicate genes; polygenic gene integration, sex determination in plants, multiple alleles, cytoplasmic inheritance, linkage and crossing over, chromosome mapping.

Unit III: Chromosomal Aberrations and Gene Mutations

Chromosomal aberrations: duplication, deletion, inversion, translocation. Mutation and its significance, addition, deletions, substitutions: transitions and transversion. Mutagens -physical and chemical agents, transposable elements in plants, DNA repair: proofreading, mismatch repair, excision repair, photoreactivation, recombinational repair and SOS- repair mechanisms.

Unit IV: Plant Breeding Principles

Importance of plant breeding in India, domestication and centers of origin of cultivated plants, plant introduction. Selection methods: mass, pure line and clonal selection. Hybridization; types, selection of parents, methods in emasculation and bagging, heterosis, genome duplication and deletion, Polyploidy and its role in plant breeding and genetic erosion, cytoplasmic molar sterility, utility in plant breeding

BTPB 0902: Plants Genomics for Crop Improvement**Unit I: Construction of Molecular Maps**

Mapping populations and near isogenic lines, different molecular marker systems for mapping, polymorphic markers and genotyping the mapping population, gene tagging and QTL. Molecular physical maps, *In situ* hybridization.

Unit II: Whole Genome Sequencing

Arabidopsis genome; Gene organization, Arabidopsis 2010 project, IRGSP project, sequencing strategy, other plant genomes and database for plant functional genomics.

Unit III: Functional Genomics

Annotations using genome, transcriptome and proteome sequences, structural and functional annotations, methods of transcriptome and proteome analysis, analysis of post-translational modification of proteins, algorithms for gene finding, annotation through homology search, ontologies for annotations, annotation databases.

Unit IV: Mutagenesis

Gene disruption, insertion and deletion mutagenesis, Molecular tags, gene entrapment and activation tagging, T-DNA insertion lines for functional analysis, transposon insertion lines for functional analysis. RNA interference; types, mechanism, and validation; virus-induced gene silencing (VIGS); vectors for VIGS and Genome editing with special reference to CRISPR-Cas.

BTPB 0903: Transgenic Plants for Crop Improvement**Unit I: Biotic Stress**

Resistance to biotic stresses; insect resistance, Bt gene and biopesticides, secondary metabolites and insect resistance. Herbicide resistance, types of herbicides, detoxification of herbicides by transgenes. Pathogen resistance; viral, fungal and bacterial resistance in transgenic plants, coat protein genes, chitinase and glucanase transgene, antimicrobial proteins.

Unit II: Abiotic Stress

Resistance against abiotic stresses; drought tolerance, DRE and DREB transcription factors, osmoprotectants, antioxidants, ion homeostasis, salinity tolerance (NHX, SOS, and HKT transporters) and acid soil tolerance (citrate and malate transporters), heavy metal tolerance; heavy metal transporters, enhancement of phytoremediation properties in transgenic plants and wasteland utilization.

Unit III: Plant Yield, Architecture and Nutritional Quality Improvement

Enhancing photosynthetic efficiency, PEPcase and IPT. Photoreceptor transgenes; Phytochrome, cryptochrome and phototropins. Nutrient use efficiency improvement; GS, GOGAT and Phot1 genes. Manipulation of architecture and flowering; PhyA, GA2, RTFL and TAC1 family genes. Genetic manipulation of flower pigmentation, anthocyanin biosynthesis genes, improvement of seed and fruit quality, TMT and FLAVR SAVR tomato and induction of early flowering

Unit IV: Molecular Farming

Transgenic plants with recombinant protein in plant root exudates; value-added special crops. Edible vaccines; selection of host plant, edible vaccine for Hepatitis B, plantibodies, production of glucocerebrosidase and hirudin.

BTPB 0904: Practical: Plant Physiology and Plant Breeding

1. Phenotypic characterization of control and drought tolerant plants.
2. Estimation of oil and fatty acids from the given seed samples.
3. Determination of osmotic potential by plasmolytic method from the given sample.
4. Determination of water potential by gravimetric method.
5. Analysis of heavy metals accumulated in given samples by atomic absorption spectroscopy.
6. Pollen germination assay by using sucrose solution.

BTPB 0905: Practical: Plant Molecular Breeding and Transgenics

1. Real-Time PCR analysis for quantification of gene expression.
2. To confirm T-DNA insertion in an Arabidopsis mutant and identify heterozygous and homozygous plants for insertion using PCR method.
3. GUS gene expression analysis by histochemical methods.
4. Analysis of the transgenic plant for the expression of foreign protein by Western blotting method.
5. Detection of transgene copy numbers by Southern analysis.
6. Intracellular protein localization by transient expression of protein: GUS/GFP Fusion constructs in onion peel cells assays by particle gun bombardment, visualization through confocal and other microscopy

SEMESTER III

BTPB 1001: Principles of Biomass and Bioenergy Sources

Unit I: Energy Sources

Fossil fuel energy and nuclear energy, non-nuclear and non-fossil fuel energy. Bioenergy-energy plantations - social forestry and silviculture energy farms.

Unit II: Biomass for Energy

Biomass materials; agricultural wastes and biomass harvesting, selection of plants with high cellulose contents, aquatic and terrestrial biomass production of algal and fungal biomass, organic wastes as a renewable source of energy and sources of wastes and composition of wastes.

Unit III: Bioenergy Sources

Petroleum plants (petrol plants); hydrocarbons for higher plants like Hevea and Euphorbia, algal hydrocarbons. Alcohols; alcohols as a liquid fuel- hydrolysis of lignocellulosic materials, ethanol production, fermentation and recovery of ethanol.

Unit IV: Biomass Conversion

Non-biological process, direct combustion (hog fuel), pyrolysis, gasification, and liquefaction. Biological process; enzymatic digestion, aerobic and anaerobic digestion

Unit V: Gaseous Fuels

Biogas and hydrogen, biogas technology, benefits from biogas plants. Biogas production: aerobic digestion solubilization, acidogenesis, methanogenesis. Biogas production from different feed stocks like *Salvinia* and *Eichhornia*. Hydrogen as a fuel: Photobiological process of hydrogen production. Hydrogenase and hydrogen production. Halobacteria.

BT 1002: Biosafety and Bioethics, Intellectual Property Rights, and Bioentrepreneurship

Unit I: Bioethics and Biosafety

Overview of research misconduct, rules and regulations in India; data management; ethical use of bioresources, transgenic crops, animal subjects; Protection of human subjects; stem cell ethics; agricultural ethics; ecosourcing-code of practice; mentor-mentee responsibilities; Authorship guidelines, Publication and peer review, plagiarism, Collaboration, Bias, Conflicts of Interest; Cyber Security Awareness; understanding phishing attacks, safe internet use, malware, antivirus software.

Chemical and biohazard safety; Social responsibility and Whistleblowing

Unit II: Essentials of Product Development:

Company protocols for research, privacy policies, institutional and professional code of ethics and standards of practice, Knowledge of basic laboratory biosafety procedures, GLP and GMP, relevant EOPs, SOPs, process flows in manufacturing, product life cycle and product properties, competitor products. Stability studies– generate stability data & prepare stability reports for innovation products

Unit III: Intellectual Property Right (IPR)

Concept and provisions of IPR; Patents, trademarks, copyright, conditional information, breeder's right. Patent; importance, types, scope, criteria, applying for a patent. Protection of biotechnological inventions.

Unit IV: Quality, Ethical and Legal Implications

International standards, quality accreditation and certification – NABH standards, Elements of quality management.

Quality checks - quality assurance samples, master sample, internal controls, statistical analysis of test data, techniques and concepts of statistical quality control and statistical process control, non-conformities. Operational aspects – calibration, accuracy checks of quality control

Privacy and confidentiality, Psychological impact, Counselling, Standards and commercialization. FDA and EPA regulations for clinical use of DNA tests and commercial release of chemical products.

Unit V: Bioentrepreneurship- trainings and workshops

A. Introduction to Bioentrepreneurship: Concepts and overview of entrepreneurship, evolution and growth of entrepreneurship in India, emerging trends in entrepreneurship development, entrepreneurial potential and potential entrepreneur, management of financing

B. Personal development and business etiquette training: Training in vital skills such as leadership skills, language and communication etiquette, dressing etiquette (for men and women), personal hygiene and cleanliness, relating with colleagues and supervisors, customer service etiquette, email etiquette, dining etiquette, telephone etiquette, training your senses, memory building, other office etiquettes.

BTPB 1003: Journal club and Group discussion

BTPB 1004: Lab Training and Dissertation on Plant Biotechnology

BTPB 1005: Industrial training

**Syllabus for M.Sc. in Biotechnology with specialization in Microbial Biotechnology
(Semester II-IV)**

Semester II				
BTMB 0801	Immunotechnology and Infection biology	T	4	50
BTMB 0802	Basics of Microbial Biotechnology	T	4	50
BTMB 0803	Environmental Microbiology	T	4	50
BTMB 0804	Practical: Immunology and Infection biology	P	4	50
BTMB 0805	Practical: Microbiology	P	4	50
Semester III				
BTMB 0901	Emerging techniques in Biotechnology	T	4	50
BTMB 0902	Food Biotechnology	T	4	50
BTMB 0903	Microbial Biomass production	T	4	50
BTMB 0904	Practical: Emerging techniques	P	4	50
BTMB 0905	Practical: Food biotechnology and Microbial biomass production	P	4	50
Semester IV				
BTMB 1001	Downstream processing	T	4	50
BT 1002*	Biosafety and Bioethics, Intellectual Property Rights and Bioentrepreneurship	T	4	50
BTMB 1003	Journal club, and Group discussion	T	4	50
BTMB 1004	Lab training and Dissertation on Microbial biotechnology	P	4	50
BTMB 1005	Industrial training	P	4	50

**This course is common for both specializations of Biotechnology*

SEMESTER II

BTMB 0801: Immunotechnology and Infection biology

Unit I: Introduction to Cellular and Molecular immunology

Fundamental concepts of the immune system: Innate immune response, adaptive immune response, B and T cell activation, complement pathway; Major Histocompatibility Complex- MHC genes, MHC and immune responsiveness and disease susceptibility, HLA typing

Advanced Immunological techniques: FACS, Immunofluorescence, Immunoblotting, ELISA

Vaccinology and Immunotherapy: Vaccine technology- Role and properties of adjuvants, recombinant DNA and protein-based vaccines; Peptide vaccines, conjugate vaccines; antibody engineering- chimeric and hybrid monoclonal antibodies; Generation of immunoglobulin gene libraries; Immunotherapy

Unit II: Microbial pathogenesis and host pathogen interaction

Mechanism of microbial pathogenesis (bacteria, virus, yeast, parasites), genetics of pathogenicity and virulence. Alteration of host cell behavior by pathogens, pathogen-induced diseases: Tuberculosis, Helicobacter, Salmonella, Hepatitis, HIV, Filariasis. Hospital-acquired infections; Pathogenic fungi; Pathogenicity of parasites (Entamoeba, Naegleria, Leishmania, Trypanosoma), mode of action, virulence

Unit III: Host-Microbiome Interaction

Microbial communities in the human body, role of Microbiota in human health; Microbial interactions with the host immune system; gut-brain axis; microbial diversity analysis; potential for microbiome-directed therapeutics to impact human disease, Metagenomics

Unit IV: Virology

Life cycle: Entry, replication and egress of DNA and RNA viruses.

Phage genetics: lytic and lysogenic cycles of bacteriophage; Virulent and temperate phage, prophage

Study of plaque morphology, mapping of phage chromosome by phage crosses.

BTMB 0802: Basics of Microbial Biotechnology

Unit I: General Characteristics, Growth and Nutrition of Microorganisms

Bacteria- Morphology and cellular component, flagella, pili, fimbriae, extracellular layers, cell wall, cell membrane, plasmids and episomes, endospore, bacterial chromosome. Extremophiles.

Growth kinetics: different phases of growth. Batch, continuous and synchronous culture. Physical factors influencing growth- temperature, pH, osmotic pressure, salt concentration.

Quorum sensing in bacteria- Role of acyl homolactone serine in quorum sensing in bacteria, types of quorum sensing, regulation of quorum sensing, cell signaling in myxobacteria.

Virus- capsid, envelope and genetic material.

General characteristics and classification of protozoa, algae, fungi.

Unit II: Control of Microorganisms

Sterilization, disinfection, antimicrobial agent (antiseptics, sanitizer, germicide, antimicrobial agent); physical method of disinfection and sterilization- dry heat, moist heat, filtration, radiation; Chemical control- dye solutions, alcohol, acid, alkali, halogen, heavy metal, phenol, phenol derivatives, formaldehyde, ethylene oxide, detergents.

Assessment of chemical disinfectant, chemotherapeutic agents- sulphonamides;

Antibiotics- mechanism of action and antimicrobial spectrum of penicillin, streptomycin, tetracycline, chloramphenicol, nalidixic acid, and metronidazole; drug resistance - phenomena and mechanism.

Unit III: General concepts of Microbial Biotechnology

General concepts of microbial biotechnology, microorganisms as source of novel compound production. Biopolymer and bioplastics, algal biotechnology, bioweapons, and bioshields. Microbes as biocontrol agents (*Baculoviruses*, *Beauveria bassiana*, *Bacillus thuringiensis*, *Bacillus sphaericus*, *Bacillus popilliae*); Microbe derived inhibitors.

Unit IV: General concepts and application of fermentation.

Fermentation- general concepts and applications; Range of fermentation process- microbial biomass, enzymes, metabolites, recombinant products, transformation process; Components of fermentation process.

Types of fermentations- aerobic and anaerobic fermentation, submerged and solid-state fermentation, factors affecting submerged and solid-state fermentation, substrates used in solid-state fermentation and its advantages; Culture media- types, components, and formulations.

Sterilization: Batch and continuous sterilization. Bioreactors, membrane Bioreactors.

Isolation, preservation, and maintenance of industrial microorganisms, kinetics of microbial growth and death, Monod model, sterilization of media for fermentation, air quality management and air sterilization. Measurement and control of fermentation parameters - pH, temperature, Oxygen requirement and supply, oxygen transfer kinetics, determination of K_La value- their effect on oxygen transfer. Effect of aeration, agitation and microbial biomass on K_La value; Newtonian and non-Newtonian fluids; Foam and antifoams.

Unit V: Process development and optimization

Process development, Optimization- classical and statistical methods of optimization; Immobilization- different matrices, whole cell, and enzyme immobilization;

Scale up of bioprocess, Analysis of batch, stability of microbial reactors, analysis of mixed microbial populations, specialized bioreactors (pulsed fluidized, photobioreactors).

Fermentation kinetics and monitoring, measurement and control of bioprocess parameters.

BTMB 0803: Environmental Microbiology**Unit I: Waste management**

Analysis of fecal indicator organisms, MPN index. BOD, COD, dissolved gases, removal of heavy metals, total organic carbon removal. Wastewater characteristics, standards of disposal, treatment objective and strategies. Treatment operations: screens, grit chambers, skimming tank, primary and secondary sedimentation tanks. Trickling filters, biofilters, rotating biological contactors.

Composting of solid wastes, aerobic & anaerobic digestion: methane production, pros and cons of anaerobic process, hydrocarbon degradation

Sludge Treatment and Disposal: Activated sludge process, oxygen and nutrient requirements, sludge thickening, design of digester tank, ultimate disposal, sludge drying beds. Oxidation ponds and lagoons.

Bioremediation- process and organisms involved; constraints and priorities of bioremediation.

Bioaugmentation; ex-situ and in-situ processes; intrinsic and engineered bioremediation.

Major pollutants and associated risks; organic pollutant degradation- microbial aspects and metabolic aspects- factors affecting the process and recent developments.

Unit II: Petroleum and Marine Microbiology

Petroleum microbiology: Types of compounds in petroleum, products of compounds in petroleum, microorganisms in hydrocarbon system, role of microorganisms in hydrocarbon degradation.

Biodegradable plastics and superbug.

Marine microbiology: Characters of the marine environment, characters of marine microorganisms, role of marine microorganisms; Microbiologically enhanced oil recovery (MEOR).

Unit III: Role of microorganisms in bioleaching and textile industry

Bioleaching of elements- introduction, microorganisms involved, chemistry of microbial leaching and beneficiation, leaching methods- laboratory and *in situ* leaching of copper and uranium.

Textile industry- introduction, types of microorganisms found on textile fibers, conditions favoring the action of microorganisms, types of destruction caused by microorganisms (cotton and wool), prevention of growth of microorganisms.

BTMB 0804: Practical: Immunotechnology and Infection biology

1. Determination of Antibody titre by ELISA
2. Isolation and purification of IgG from serum
3. To detect a specific antigen in a cell homogenate using Western blot hybridization.
4. Determination of viral titre by plaque assay
5. Immunofluorescence observation of subcellular structures of mammalian cells

BTMB 0805: Practical: Microbiology

1. Aseptic techniques in Microbiology
2. Characterization of bacteria from environmental samples (sample collection, serial dilution, media preparation plating, pure culture preparation, staining and biochemical tests)
3. Microbiological assay of antibiotics (MIC and Paper disc)

4. Testing the samples for BOD, COD, TDS.
5. Bacteriological Examination of Water, MPN and IMVIC Test.
6. Monitoring of pollution indicating microbe communities in case of hydrocarbon-polluted soil systems. Interpretation of germ count/activity results.

SEMESTER III

BTMB 0901: Emerging techniques in Biotechnology

Unit I: Recombinant DNA technology

Restriction endonucleases, restriction mapping, DNA and RNA modifying enzymes (polymerase, reverse transcriptase, ligase, alkaline phosphatases, terminal transferase, nuclease)

Vectors (plasmids, cosmids, bacteriophages, phagemid, BAC and YAC, shuttle vectors).

Cloning methods (directional and gateway), introducing engineered plasmids into a bacterial cell (transformation, conjugation, and transduction).

Design, use and properties of expression systems; Yeast two-hybrid systems; baculovirus and *Pichia* expression systems; regeneration of active proteins from inclusion bodies; Phage display

Construction of cDNA and genomic DNA libraries, screening of libraries, use of linker and adaptors.

Identification and analysis of recombinant DNA clones, use of transposon in genetic analysis. Genetic manipulation of microorganisms and strain improvement.

Unit II: Techniques in genetic engineering

Types of PCR- (Asymmetric, multiplex, nested, hot start, touch up, touchdown, RT-PCR, overlap extension, SOEing, and site-directed mutagenesis). PCR in molecular diagnostics- detection of viral diseases, PCR in detecting gene disorder and minimum residual disease.

Methods of nucleic acid detection, RFLP, DGGE, DNA-protein interaction study-EMSA, DNA footprinting; S1 nuclease mapping, RNase protection assay; strategies of gene delivery (chemical, physical or mechanical method), Genome editing tools.

Unit III: Biophysical techniques

Absorption, fluorescence and emission spectroscopy of biomolecules.

Centrifugation: Principle of centrifugation and different types of centrifuges; differential & density gradient centrifugation

Circular dichroism, Fluorescence Anisotropy, Isothermal Calorimetry; Structure determination of biomolecules using NMR spectroscopy and X-ray crystallography.

Unit IV: Genomics

Basics of genomics; Genome mapping; markers for genetic mapping; methods and techniques used for gene mapping, physical mapping, linkage analysis, cytogenetic techniques, Fluorescent In Situ

Hybridization in gene mapping, somatic cell hybridization, radiation hybrid maps

Genome sequencing projects for microbes, plants and animals; Human Genome Project, accessing and retrieving genome project information from the web. Next-generation sequencing
Comparative genomics- identification and classification of organisms using molecular markers- 16S rRNA typing/ sequencing, SNPs; comparative gene mapping

Unit IV: Proteomics

Functional genomics, proteomics, and concepts of proteogenomics, overview of technologies and challenges to study systems at different levels; isoelectric focusing and 2D-PAGE, DIGE, mass spectrometry, protein and peptide sequencing; Quantitative proteomics-SILAC
Application of proteomics

Unit V: Radiolabeling techniques

Properties of different types of radioisotopes normally used in biology, their detection and measurement, incorporation of radioisotopes in biological tissues and cells, molecular imaging of radioactive material, safety guidelines- radioactive waste management and disposal

Unit VI: Nanotechnology

Elementary concept of nanotechnology and its applications; cellular nanomachines; bio-inspired nanomaterials for a new generation of medicine; nanoscience in medicine, delivery system, and vaccine; nanoparticles in diagnostics; synthesis of inorganic/organic nanoparticles and characterization.

BTMB 0902: Food Biotechnology

Unit I: Microbes and Food Spoilage

Importance of microbes in food, sources of microbes in food, normal microbiological quality of food, factors influencing microbial growth in food. Sources of food contamination. Principles of food spoilage; spoilage of cereals, sugar products, vegetables, fruits, meat, and meat products, milk and milk products, fish and sea foods, poultry; spoilage of canned foods; conventional and modern methods for detection of spoilage and characterization.

Unit II: Foodborne Infections and Intoxication

Food infections (sources, transmission, and control) by bacteria- Brucella, Bacillus, Clostridium, Escherichia, Listeria.

Food intoxication (sources, transmission, and control)- Botulism, Staphylococcal

Mycotoxins & their types- aflatoxins, ochratoxins, fumonisins, trichothecenes, zearalenone, ergot alkaloids; food borne outbreaks and lab testing procedures. Preventive measures. Molds, Algae, Protozoa, Viruses.

Unit III: Food Preservation

Principles and methods of food preservation- physical (temperature, irradiation, drying, canning, processing for heat treatment-D, Z and F values), modifications of atmosphere, control of water activity, compartmentalization; Chemical (Organic acids, food additives. class I and class II preservatives); Control by combination of methods (Hurdle concept), biopreservation (lactic acid bacteria). Food packaging- types of packaging materials, properties, and benefits, Canning.

Unit IV: Uses of Microbes in Food

History, scope and importance of fermented foods; Organisms used for production of fermented food products. Microbial stress response in food, starter cultures, microbiology of fermented foods. General method of production, fermented vegetables (soy product, sauerkraut), meat (sausages), beverage (cocoa and coffee); Bread, idli, dairy foods (cheese, shrikhand). Production methods of Kefir, Yogurt, Acidophilus milk; Probiotics, prebiotics and synbiotics, nutraceuticals (Cr/Se yeast), functional foods and their quality standards. Application of fungal pigments in the food industry, SCP- Nutritional & therapeutic importance, Quorn and SCO and their Industrial production.

Unit V: Microbial Detection and Food Safety

Conventional Methods, sampling for microbial analysis, quantitative microbial enumeration in food, qualitative methods of microbial detection, rapid methods- biosensors, controlling the microbiological quality of food, quality and criteria, sampling schemes, Good Hygiene Practices, sanitation in manufacturing and retail trade; food control agencies and their regulation, QC using microbiological control, control at source, codes of GMP, HACCP, DNA barcoding, laboratory accreditation

BTMB 0903: Microbial Biomass production

Unit I: Primary and secondary metabolite production

Microbial products as primary and secondary metabolites; trophophase-idiophase relationships in production of secondary metabolite; Role of secondary metabolites in physiology of organisms producing them; Pathways for synthesis and control of primary and secondary metabolites of commercial importance.

Unit II: Production of Microbial Biomass

Production of Microbial Biomass - Baker's yeast, mushroom; Production of fermented foods; Alcoholic beverages- wine, beer, etc; Production of ethanol, citric acid; amino acids and vitamins; microbial enzymes for food, detergent and pharma industry; biopesticides and biofertilizers
 Production of antibiotics; Penicillin and other antibiotics; pigments, microbial transformation, production of Insulin, Interleukin, growth hormones, etc using recombinant DNA technology.

Unit III: Microbial inoculants

Microbial inoculants- Selection and establishment of nitrogen-fixing bacteria. Production of *Rhizobium*, *Azotobacter*, *Azospirilla*, *Azolla*, cyanobacteria and other nitrogen-fixing bacterial cultures.

Quality control of bio inoculants; Phosphate solubilizing bacteria; mycorrhiza; plant growth promoting rhizobacteria (PGPR); Composting and biocomposting, biocontrol microbial inoculants.

Unit IV: Usage of microorganisms in industries

Use of microorganisms in producing biomaterials, restriction enzymes, cyanobacterial cultures for Polyhydroxyalkanoates (PHAs); Microbial synthesis of nanoparticles and their applications, Bio-mineralisation by microorganisms.

Use of fungi in industry- a) Food industry, biosensors and fuel cells (architecture of the fungal cell: cell wall, membranes and cytoskeleton); b) use of fungi in agriculture and environmental applications; c) Biofertilizers, bioremediation and biological control, and iv) Food industry, biosensors, and fuel cells

BTMB 0904: Practical: Emerging techniques

1. PCR amplification of a DNA fragment, quantitation by PCR
2. Restriction Enzyme digestion of DNA and gel electrophoresis
3. Gene cloning and recombinant screening
4. Confirmation of the insert by Colony PCR and restriction mapping
5. Expression of tagged recombinant protein in *E. coli* and purification by affinity chromatography
6. Green synthesis of nanoparticles

BTMB 0905: Practical: Food biotechnology and Microbial biomass production

1. Detection and enumeration of indicator and index microorganisms for food borne pathogens (total enterobacteria, total coliform & aerobic spore former)
2. Identification of spoilage causing bacteria and fungi of food samples – fruits, vegetables, bread.
3. Enrichment and isolation of N₂-fixing bacteria from plant associated niche.
4. Laboratory fermenter sterilization, operations and scale up of selected strain.

5. Isolation of lactic acid producing bacteria and production of fermented milk products/
Sauerkraut.
6. Characterization of cellulose/ pectin decomposition, starch hydrolyzing microorganisms
from environmental samples.
7. Preparation of spawn for mushroom cultivation

SEMESTER IV

BTMB 1001: Downstream Processing

Unit I: Necessity of Downstream Processing

Overview of a bioprocess including upstream and downstream processing; Importance of downstream processing in biotechnology, characteristics of biological molecules and their separation characteristics based on stability & other biological properties, problems and requirements of bioproduct purification; Characteristics of biological mixtures; Downstream process economics.

Unit II: Biomass Removal and Cell Disruption

Physico-chemical basis of bioseparation processes. Removal of particulate matter; biomass; and insoluble; flocculation and sedimentation, filtration methods, application of centrifugation
Cell disruption- mechanical, enzymatic, and chemical methods.

Unit III: Membrane-Based Separation and Chromatography

Membrane separations: Membrane-based separation theory; types of membranes; Types of membrane processes (Dialysis; ultrafiltration; microfiltration; Reverse Osmosis).
Chromatography Techniques- TLC, HPLC, HPTLC & FPLC, Size-exclusion Chromatography, Affinity chromatography, Ion-exchange Chromatography; Gas chromatography, electrophoretic separation.

Unit IV: Product Polishing, Crystallization, Drying and Case Studies

Final product polishing and case studies, products polishing; crystallization and drying; Formulation, purification of antibiotics, microbial enzymes, exopolysaccharides, use of immobilized cells/enzymes. CIPP/RIPP schemes for ethanol, methanol, citric acid.

BT 1002: Biosafety and Bioethics, Intellectual Property Rights, and Bioentrepreneurship

Unit I: Bioethics and Biosafety

Overview of research misconduct, rules and regulations in India; data management; ethical use of bioresources, transgenic crops, animal subjects; Protection of human subjects; stem cell ethics; agricultural ethics; ecosourcing-code of practice; mentor-mentee responsibilities; Authorship guidelines, Publication and peer review, plagiarism, Collaboration, Bias, Conflicts of Interest; Cyber Security Awareness; understanding phishing attacks, safe internet use, malware, antivirus software.
Chemical and biohazard safety; Social responsibility and Whistleblowing

Unit II: Essentials of Product Development:

Company protocols for research, privacy policies, institutional and professional code of ethics and standards of practice, Knowledge of basic laboratory biosafety procedures, GLP and GMP, relevant EOPs, SOPs, process flows in manufacturing, product life cycle and product properties, competitor products. Stability studies– generate stability data & prepare stability reports for innovation products

Unit III: Intellectual Property Right (IPR)

Concept and provisions of IPR; Patents, Trademarks, Copyright, Conditional information, Breeder's right. Patent; importance, types, scope, criteria, applying for a patent. Protection of Biotechnological inventions.

Unit IV: Quality, Ethical and Legal Implications

International standards, Quality accreditation and certification – NABH standards, Elements of quality management.

Quality checks - quality assurance samples, master sample, internal controls, statistical analysis of test data, techniques and concepts of statistical quality control and statistical process control, non-conformities. Operational aspects – calibration, accuracy checks of quality control

Privacy and confidentiality, Psychological impact, Counselling, Standards and commercialization. FDA and EPA regulations for clinical use of DNA tests and commercial release of chemical products.

Unit V: Bioentrepreneurship- trainings and workshops

A. Introduction to Bioentrepreneurship: Concepts and overview of entrepreneurship, evolution and growth of entrepreneurship in India, emerging trends in entrepreneurship development, entrepreneurial potential and potential entrepreneur, management of financing

B. Personal development and business etiquette training: Training in vital skills such as leadership skills, language and communication etiquette, dressing etiquette (for men and women), personal hygiene and cleanliness, relating with colleagues and supervisors, customer service etiquette, email etiquette, dining etiquette, telephone etiquette, training your senses, memory building, other office etiquettes.

BTMB 1003: Journal club and Group discussion

BTMB 1004: Lab Training and Dissertation on Microbial biotechnology

BTMB 1005: Industrial training