

THIRD SEMESTER

Course No.-IX: Environmental Biotechnology

UNIT-I

Environment: Basic concepts and issues; environmental pollution: types and methods for the measurement, Impacts of Environmental Pollution at local, regional and global level. air pollution and its control through biotechnology, air sampling techniques, Biodiversity: conservation and management and Introduction to environment Law

UNIT-II

Water pollution and its control: Water monitoring and water quality guidelines, waste water treatment: Aerobic process-activated sludge, oxidation ditches, trickling filter, towers, rotating discs, rotating drums, oxidation ponds; anaerobic processes-anaerobic digestion, anaerobic filters, upflow anaerobic sludge blanket reactors; treatment schemes for waste waters of dairy, distillery, tannery industries; biotechnological application of microbes from extreme environment, algal blooms and human health.

UNIT-III

Solid Waste management: Concept of solid waste; types of solid waste, Major sources of solid wastes; Effects of solid waste generation on quality of air, water and public health; Disposal of plastic waste and hazardous wastes,; Disposal of medical waste, Biotechnological approaches for solid waste management.

UNIT-IV

Microbial degradation of xenobiotics in the environment- ecological considerations, decay behaviour & degradative plasmids, hydrocarbons, substituted hydrocarbons, oil pollution, surfactants, pesticides, fertilizers; bioaccumulation of metals and radio-nucleids and detoxification; bioremediation.

UNIT-V

Biological N₂ fixation, H₂ production, biofertilizers and biopesticides; solid wastes; sources and management (composting, vermiculture and methane production). Single cell protein (Spirulina, yeast, mushroom); global environmental problems-ozone depletion, UV-B, green house effect and acid rain, their impact and biotechnology approaches for management. Basic idea of Organic farming.

Practical

1. To detect coliforms for determination of the purity of potable water.
2. To determine dissolved oxygen concentration of water sample.
3. To determine biological oxygen demand (BOD) of a sewage sample.
4. To estimate nitrate, nitrite, and ammonium in drinking water.
5. To study the impact of heavy metals on growth & survival of microbes.
6. To study the impact of pesticides on the growth and survival of microbes.
7. To study the impact of salt and osmotic stress on the growth and survival of microbes.

Books

1. Metcalf and Eddy (1999). Wastewater Engineering- Treatment, disposal and Reuse. Inc., Tata McGraw Hill, New Delhi.
2. Moo-Young, M., Comprehensive Biotechnology. (Vol- 4), Pergamon Press, Oxford.
3. De, A.K. Environmental Chemistry, Wiley Eastern Ltd., New Delhi.
4. Allsopp, D. and K.J. Seal (1986) Introduction to Biodeterioration. ELBS/Edward Arnold, London.
5. Thakur, I.S. Environmental Biotechnology – Basic concepts and applications (2006) , I K International publications

Course No.-X: Genetic Engineering

UNIT-I

Scope of genetic engineering, milestones in genetic engineering; RDT:-Basic concept, cloning and gene expression; cloning and patenting of life forms; genetic engineering guidelines; molecular tools and their applications; restriction enzymes, modification enzymes, DNA and RNA markers; nucleic acid isolation and purification, yield analysis.

UNIT-II

Nucleic acid amplification: PCR and its applications; gene cloning vectors: plasmids, bacteriophages, phagemids, cosmids, artificial chromosomes; restriction mapping of DNA fragments and map construction; nucleic acid sequencing; cDNA synthesis and cloning; reverse transcription, DNA primers, linkers, adaptors and their chemical synthesis; library construction and screening.

UNIT-III

Alternative strategies of gene cloning:- cloning of interacting genes: two-and three hybrid systems, cloning of differentially expressed genes ;Nucleic acid micro array; site-directed mutagenesis and Protein engineering; Northern blot; primer extension; S1 mapping; RNase protection assay; reporter assays, SNP Mapping.

UNIT-IV

Expression strategies for heterologous genes: vector engineering and codon optimization, Host engineering; In vitro transcription and translation, expression of proteins in bacteria, yeast, insects, mammalian, and plant cells; processing of recombinant proteins: purification and refolding, characterization of recombinant proteins, stabilization of proteins; phage display.

UNIT-V

T-DNA and transposon tagging:-role of gene tagging in gene analysis, identification and isolation of genes through T-DNA or transposon; transgenic and gene knockout technologies: targeted gene replacement, chromosome engineering; gene therapy; vector engineering, strategies of gene delivery, gene replacement/gene correction/ gene editing, gene regulation and silencing.

Practical

1. To study Isolation of antibiotic resistant bacteria from natural sources.
2. To study Preparation of competent cells.
3. To isolate plasmid DNA.
4. To study Quantitation of nucleic acids.
5. To study Restriction mapping of DNA.
6. To study Construction of restriction map of plasmid DNA.
7. To study amplification of DNA fragments by PCR and comparison with standard DNA.

Books

1. J. Sambrook , E. F. Fritsch and T. Maniatis, Molecular cloning: A Laboratory Manual,. Cold Spring Harbor Laboratory Press, New York.
2. D. M. Glover and B. D. Hames, DNA Cloning: A practical Approach, IRLPress, Oxford,
3. P. B. Kaufman, W. Wu. D. Kim and L. J. Cseke, Molecular and Cellular Methods in Biology and Medicine, CRC Press, Florida,
4. S.L. Berger and A. R. Kimmel, Methods in Enzymology (Vol. 152), Guide to Molecular Cloning Techniques, Academic Press, Inc. San Diego,
5. D.V. Goeddel, Methods in Enzymology (Vol.185) Gene Expression Technology, Academic Press, Inc. San Diego.
6. D. A. Mickloss and G. A. Freyer, DNA Science. A First Course In Recombinant Technology, Cold Spring Harbor Laboratory Press, New York,
7. S.B. Primrose, Molecular Biotechnology (2nd Edn.), Blackwell Scientific Publishers, Oxford.
8. J. A. Davies and W. S. Reznikoff. Milestones in Biotechnology, Classic Papers in Genetic Engineering, Butterworth- Heinemann, Boston,
9. M. R. Walker and R. Rapley. Route Maps in Gene Technology, Blackwell Science Ltd. Oxford
10. S. M. Kingsman and A. J. Kingsman. Genetic Engineering; An Introduction to gene analysis and exploitation in eukaryotes, Blackwell Scientific Publications, Oxford.
11. B. J. Glick, J. J. Pasternak and Patten C. L., Molecular Biotechnology: Principles and Application of Recombinant DNA, American Society for Microbiology Press

Course No. XI: Plant Biotechnology**UNIT-I**

Introduction to plant cell and tissue culture: tissue culture media (composition and preparation), initiation and maintenance of callus and suspension culture. Regeneration through organogenesis and somatic embryogenesis; transfer and establishment of whole plant in soil; embryo culture and embryo rescue; anther, pollen and ovary culture for production of haploid plants and homozygous diploid lines; Cryopreservation for germplasm conservation; protoplast isolation, culture and fusion; Selection of hybrid cells and regeneration of hybrid plant; symmetric and asymmetric cybrids; germplasm conservation, virus free plants.

UNIT-II

Cloning vectors for transformation in higher plant: *Agrobacterium tumefaciens*, Ti and Ri plasmids, basis of tumor formation, hairy root, mechanisms of DNA transfer, role of virulence genes. Viral vectors and their application, direct gene transfer: particle bombardment, electro poration, microinjection. Transformation of monocots; transgene stability and gene silencing, selection of clones. Expression of cloned genes: genetic markers, reporter genes, Gus assay.

UNIT-III

Application of plant transformation for productivity and performance: herbicide resistance (phosphinothricin, glyphosate, sulfonylurea, atrazine), insect resistance (Bt. endotoxin genes, Non-Bt like proteinase inhibitors, alpha amylase inhibitor), virus resistance (Coat protein mediated protection (CPMP), nucleocapsid gene), Disease resistance (anti fungal proteins: chitinase, 1-3 beta glucanase), ribosome inactivating proteins (RIP), thionins, pathogenesis related (PR) proteins, nematode resistance, abiotic stress (salt tolerance); post harvest losses, long shelf life of fruits and flowers, use of ACC synthase, polygalacturanase, ACC oxidase, carbohydrate composition and concentration during storage. ADP glucose pyrophosphatase.

UNIT-IV

Chloroplast transformation: advantages, vectors, success with tobacco and potato; Metabolic engineering and industrial products; plant secondary metabolites, control mechanism and manipulation of phenyl propanoid pathway, Shikimate pathway, alkaloids, industrial enzymes; biodegradable plastics. Polyhydroxybutyrate, therapeutic proteins; lysosomal enzymes, antibodies, edible vaccines, purification strategies: oleosin partitioning technology.

UNIT-V

Molecular markers related to plant breeding: RFLP maps, Linkage analysis, RAPD markers, STS, microsatellites, SCAR (sequence characterized amplified region), AFLP, QTL of crops with suitable examples. Molecular assisted selection; arid and semi- arid plant biotechnology, green house and green- home technology, Concept and Applications of Hydroponics

Practical

1. To prepare media for plant tissue culture.
2. To perform surface sterilization of explant.

3. To perform organ culture of given explants.
4. To study Regeneration of whole plant through Callus culture.
5. To isolate and culture protoplast from given plant cell.
6. To perform Anther culture for production of haploid plants.
7. To isolate DNA from plant leaves.

Books

1. Hammond J., McGarvey P. and Yusibov V. (Eds.), Plant Biotechnology. SpringerVerlag,
2. Fu T.J., Singh G. and Curtis W.R. (Eds). Plant Cell and Tissue Culture for the Production of Food Ingredients, Kluwer Academic/Plenum Press.
3. Chawla H.S Plant Biotechnology, International Book Distributing Company.
4. Henry R.J. Practical Application of Plant Molecular Biotechnology. Chapman and Hall.
5. Gupta P.K. Elements of Biotechnology. Rastogi and Co. Meerut.

Course No. XII: Bioprocess Engineering and Technology

UNIT-I

Biofermentation: Designing and application, Types of biofermentation, monitoring and control of parameters (pH, oxygen, agitation, temperature, foam etc.), Fermentation Kinetics (product yield, biomass yield and specific growth rate), batch & continuous culture; production medium, raw materials, isolations; maintenance, preservation & improvement of industrial strains.

UNIT-II

Upstream processing methods, Downstream processing: Filtration of fermentation broths, ultra-centrifugation, recovery of biological products by distillation, superficial fluid extraction. Comparison of Up-stream processing and Downstream processing methods

UNIT-III

Industrial production of solvents: Ethyl alcohol, citric and acetic acids; enzymes; amylases, proteases, cellulases; vitamins: vitamin B₁₂, vitamin C, antibiotics (penicillin, streptomycin, tetracycline and griseofulvin). Microbes in petroleum industry (oil recovery); immobilized cells & enzymes.

UNIT-IV

Microbes in food processing: Microbes and their use in pickling, producing colours and flavours, alcoholic beverages and other products; Processing wastes-whey, molasses, starch substrates and other food wastes for bioconversion to useful products

Dairy microbiology: Microbial examination of milk: pasteurization and phosphatase test, sterilization of milk, grades of milk, dairy products, fermented milk, butter & cheese.

UNIT-V

Packaging Technology: Definitions, functions of packaging. Properties of packaging material in relation to these functions, package design, Tests for flexible packaging materials, different levels of packaging, materials used in packaging, types of containers-primary & secondary, flexible & rigid, hermetic & non hermetic

Practicals

1. To isolate industrially important microorganisms for microbial processes.
2. To determine thermal death point (TDP) and thermal death time (TDT) of microorganism.
3. To study cell immobilization using alginate method
4. To isolate antibiotic producing microbes..
5. To perform quantitative test of milk by resazurin test.
6. To isolate fungi from spoiled bread

Books

1. Aiba S., Humphrey A.E. and Millis N. F. Biochemical Engineering, Univ of Tokyo Press, Tokyo.
2. Atkinson B. Biochemical Reactors, Pion Ltd. London.
3. Baily, and Ollis, Biochemical Engineering Fundamentals, McGraw-Hill Book Co. New York.

4. Stockholm. Bioprocess Technology: Fundamental and Application,
5. Jackson, Process Engineering in Biotechnology, Prentice Hall, Engelwood Cliffs.
6. Shuler and Kargi, Bioprocess Engineering: Basic Concepts, Prentice Hall, Engelwood Cliffs.
7. Stanbury and Whitaker, Principles of Fermentation Technology, A. Pergamon Press, Oxford.
8. Nielson and Billadsen, Bioreaction Engineering principles, J. Plenum Press.
9. Shuler, Chemical Engineering Problems in Biotechnology, (Ed.) AICHE.
10. J.M. Lee, Biochemical Engineering, Prentice Hall Inc.
11. W.F.Viet, Bioprocess Engineering-kinetics, Mass Transport, Reactors and Gene Expression, John Wiley & Sons, Inc.