

## SEMESTER IV

### Paper 13: Molecular Phylogeny and Extremophiles (GMBT-401)

#### Course Objective:

"At the end of this course the students will be able to describe, the student will understand the Extremophiles Help Us Predict the Boundaries of Life Over the past century, the boundary conditions under which life can thrive have been pushed in every possible direction, encompassing broader swaths of temperature, pH, pressure, radiation, salinity, energy, and nutrient limitation."

#### Course Contents:

##### UNIT-1: Microbial evolution and phylogeny

Molecular basis of microbial classification, Chronometers and chronological distances, Paradox in establishing Evolutionary distances.

##### UNIT-2: Non-cultivable microbes

Cultivable vs. non-cultivable microbes, Genetic heterogeneity among non-cultivable, Metabolic potential of non-cultivable microbes, Evolutionary and biotechnological significance of non-cultivable microbes

Molecular methods for studying non-cultivable microbes: Isolation of nucleic acids and analyses of microbial diversity, In-situ hybridization, Methods of 16S rRNA analysis.

##### UNIT-3: Archaeobacteria:

Archaeobacteria - distinguishing features, Phylogenetic groups of Archaeobacteria,

Ecology and habitats of Archaeobacteria, Physiology of Archaeobacteria

##### UNIT-4: Life at Extremities:

Life at hyper-extremities: hyperthermophilic Archaeobacteria and bacteria, Life at hyper salinity, other forms of extremophiles Gene expression in hyperthermophilic bacteria and archaea, Genome analysis from extremophiles Protein stability in hyper-extremophiles

##### UNIT-5: Biotechnology and Extremophiles:

Hyper-extremophiles and their novel metabolic machinery and biomolecules- future unique applications

#### RECOMMENDED BOOKS:

1. Henderson et al.(1999). Cellular Microbiology.Wiley.
2. De Bruijn et al.(1998). Bacterial Genomes.Chapman & Hall.
3. Goldsby, Kindt & Osborne(2006). Kuby's Immunology WH Freeman & co.
4. Tizard(1995). An Introduction to Immunology.Harcourt Brace College Publication
5. Delves PJ & Roitt IM(2006). Roitt's Essential Immunology, 11th ed., Wiley-Blackwell

#### Practicals

1. Estimation of organic matter in agricultural soils to assess the soil fertility.
2. Estimation of cell wall degrading enzymes : cellulases (exo-and endo glucanases), polymethyl esterase, poly galacturonase, pectic lyase in host-pathogen interactions
3. Estimation of accumulated soil enzymes : catalase/peroxidase, phosphatase, urease,
4. Isolation and identification of cyanobacteria used as biofertilizers- *Nostoc*, *Anabaena*, *Scytonema*
5. Isolation of *Rhizobium* from root nodules
6. Classification and symptomatology of plant diseases covered in theory (unit III)
7. Determination of Disease Tolerance Index (DTI) in crop plants
8. Biochemical changes in healthy and diseased crop plants : carbohydrates, proteins, amino acids, chlorophyll
9. Quantification of phytoalexins in healthy and diseased crop plants
10. Analysis of PR proteins in healthy and diseased plants through electrophoresis

### Paper 14: Molecular Biotechnology (GMBT-402)

#### Course Objective:

This course is designed as such to target the knowledge and understanding of Nucleic acids and the experiments related to Genetics. After successful completion, this course will give the students in-depth knowledge about the prokaryotic and eukaryotic DNA replication and chemistry of DNA synthesis. This course will enhance the students in investigation of genetic

principle, DNA priming and DNA replication enzymes. On completion of this course, the students will gain an understanding of the structure of DNA and RNA, their types and organization. This course will gain an insight into the genomic structure, Nucleosome, Chromatin structure and organization of chromosomes."

### **Course Contents**

#### **UNIT – 1: Proteomics techniques**

Techniques used in gene detection and expression studies: Southern hybridization, Northern hybridization, western hybridization, PCR and RT-PCR

Peptide sequencing and synthesis: principles and strategies for protein sequencing. Design of primers from amino acids sequences.

#### **UNIT – 2: DNA- protein interaction techniques**

Gel mobility shift assay, DNA-protein cross-linking assay, Dnase I foot printing and SI nuclease mapping. Protein-protein interactions: chemical cross-linking. Yeast-2-hybrid, Yeast-3-hybrid and their various versions. Principles and applications.

#### **UNIT – 3: Reporter genes**

Chloramphenicol acetyl transferase (*cat*), neomycin phosphoryl transferase II (*nptII*), Luciferase,  $\beta$ -galactosidase etc. and their applications in expression kinetics and promoter probing studies.

#### **UNIT – 4: Protein folding**

Protein folding and the roles of Molecular chaperones, Mechanism and relevance to biotechnology, Assisted protein folding, *In-vitro* protein folding

#### **UNIT – 5: Protein engineering and drugs design**

Rational of protein engineering, Methods and approaches: directed Evolution and gene shuffling, random mutagenesis and selection of engineered proteins, gene modification at specific sites, synthesis of complete gene. Engineering by gene fusion. Drug design and various approaches: by blocking enzyme activity, Inhibitor for Dihydroxyfolate reductase (DHFR), Renin. HIV reverse transcriptase etc Drug design by blocking hormone receptors, propranolol for norepinephrine and epinephrine etc, and drug design by inhibiting nucleic acid synthesis using antisense RNA technology.

### **Recommended Books:**

1. Molecular Biology by David P. Clarke, 1st edition; Elsevier Academic Press; 2005.
2. Molecular Cloning: A laboratory manual by Joseph Sambrook & David Russell, 3rd edition; CSHL Press; 2001.
3. DNA Technology : The Awesome Skill by I. Edward Alcamo, 2nd edition; Hardcourt Academic Press; 2001.
4. Molecular Biology of the Gene by James Watson, Tania Baker, Stephen Bell, Alexander Gann, Michael Levine & Richard Losick, 6th Edition; CSHL Press; 2007.

### **Practicals**

1. Isolation of Genomic DNA from a bacterium and its quantification.
2. Designing primers for 16S rRNA gene sequence.
3. Amplification of 16S rRNA gene sequences by using genomic DNA as well as by colony boiling method.
4. Purification of 16S rRNA gene.
5. Sequence of 16S rRNA gene; editing the sequence, multiple alignments, construction of phylogenetic trees and interpretation of results.
6. Dot blot hybridization of different eubacterial species and interpretation of results.

### **ELECTIVE PAPER (ANY ONE)**

#### **Paper 15: Pharmaceutical Biotechnology (GMBT-403)**

##### **Course Objective**

"This course is designed as such to target the knowledge and understanding Pharmaceutical biotechnology is a relatively new and growing field in which the principles of biotechnology are applied to the development of drugs. A majority of therapeutic drugs in the current market are bioformulations, such as antibodies, nucleic acid products and vaccines. On completion of

this course, the students will gain an understanding of the structure of DNA and RNA, their types and organization."

### **Course Contents**

#### **UNIT – 1: Structural and functional genomics**

Structural and functional organization of the human genome.

Physical mapping and linkage analysis, Identification of the disease linked genes and markers, positional cloning, isolation of the disease responsible genes and their characterization. Global genome functional variations: assessment by microarrays (cDNA and Oligo microarrays), 2D protein gel electrophoresis, MALDI. Functional analysis of human genome for studying the diseases and drug functionality and drug side effects (by microarray and 2D gel electrophoresis).

#### **UNIT – 2: AIDS**

History of HIV, types, Life cycle to the HIV, Genome variations among the HIV strains, Key aspects for the drug designing targets.

#### **UNIT – 3: Pharmacogenomics and Molecular Diagnostics**

Importance and types of drug metabolizing enzymes, Variations in the drug metabolizing genes their effects, Individualized medicine and their, application in the drug dosage and treatment in cancer. Principles and application of the molecular diagnosis via protein, DNA and other biomolecular detections.

#### **UNIT – 4: Antibiotics and Pharmacokinetics**

Antimicrobial agents, Vaccines, Modern approaches in Vaccination, physiologic and biochemical processes influence the fate of drugs in the body. The interrelationship between the physicochemical properties of the drug and the rate/extent of absorption. Fundamental pharmacokinetic principles and quantitative relationships, evaluating pharmacologic response and explaining mechanisms of drug-drug interactions.

#### **Recommended Books:**

1. Peruski, L.F. Jr. and Peruski, A.H. (1997). The Internet and New Biology: Tools for Genomic and Molecular Research ASM.
2. Schena, M. Ed. (1999). DNA Microarrays: A Practical Approach. Oxford University Press.
3. Hunt, S. and Livesey, F. Ed. (2000). Functional Genomics: A Practical Approach. Oxford University Press.
4. Recent Articles in Journals.
5. Twyman (2004). Principles of Proteomics.
6. Lieblker, D.C. (2007). Introduction to Proteomic: Tools for the New Biology.

#### **Practicals**

1. Isolation of amylase producing bacteria from soil
2. Preparation of standard curve of reducing sugars by DNS method
3. Quantitative estimation of amylolytic potential of isolated bacterial culture
4. Isolation of yeasts from natural environment
5. To perform an experiment to show the Ethanol fermentation by yeast.
6. Quantitative estimation of ethanol by distillation method
7. Demonstration of surface fermentation
8. To isolate plasmid DNA from a given culture
9. To prepare agarose gel and to run the plasmid DNA samples
10. Isolation of chromosomal DNA
11. Plant DNA extraction by Phenol: Chlorophorm method
12. Demonstration of submerged fermentation
13. Demonstration of solid state fermentation

### **Paper 15: Agricultural Biotechnology (GMBT-403)**

#### **Course Objectives**

The student will be able to understand the basic techniques in animal biotechnology to improve yield of the plants. (ii) After finishing this course, the student will learn the genetic manipulation technique for crop improvement. (iii) The student will understand the application of microbial agents in agrochemical industries. (iv) The student will learn the advancement in genetic engineering techniques in agriculture. (v) At the end of this course, the student will learn the use of biofertilizers and ethical aspects in agriculture and animal farming."

### **Course Contents**

**UNIT – 1: Taxonomy and physiology**

Classification of plant kingdom (Bentham and Hooker) Absorption of water, mineral nutrition, transpiration, phytohormones

**UNIT – 2: Molecular Biology of Stress Tolerance in Plants**

Basic plant physiology and regulation Water Stress, salt stress, High Temperature Stress, Freezing Stress, Systems Biology to Study Cold Tolerance, Nutrient Stress, Heavy Metal Stress

**UNIT – 3: Genetic Transformation of Plants**

Agrobacterium mediated and biolistics-basic principles and applications, Ti plasmids, binary vectors, transformation hosts, Selection markers, Reporter genes, promoters Mechanism of transformation Screening of the transgenic plants and heterologous gene expression

**UNIT – 4: Molecular farming: (Reported examples)**

Transgenic crop with Heat shock proteins, Ion/proton transporters, Reactive oxygen scavenger, Transcription and factors Transgenic plants with pathogenetic resistance protein. Plant-derived recombinant therapeutic protein, plant-derived recombinant antibody, vaccine candidate –hepatitis B virus surface antigen in tobacco, plant-derived industrial enzyme, amylase in tobacco, Secretory IgA produced in tobacco

**Recommended Books:**

1. Bajaj, Y.P.S. (Ed.). Biotechnology in agriculture and forestry. Various volumes published time to time. Springer-Verlag, Berlin
2. Bhojwani, S.S. 1990. Plant tissue culture: Applications and limitations. Elsevier Publishers, Amsterdam.
3. Bhojwani, S.S., and Razdan, M.K. 1996. Plant tissue culture: Theory and Practice. Elsevier Publishers, Amsterdam.
4. Dixon, R.A. and Gonzales, R.A. (Ed.) 1994. Plant cell culture, a practical approach. 2nd edition. Oxford University Press, Oxford. U.K.
5. Evans, D.A., Dharp, D.R., Ammirato, P.V. and Yamuda, Y. (Ed.). Handbook of Plant cell culture series. Vol. 1-6, McGraw Hill Publishing Company, New York.
6. Gamborg, O.L. and Phillips, G.C. 1995. Plant cell, tissue and organ culture, fundamental methods. Springer International student edition. New Delhi.
7. George, E.F. 1993 / 1996. Plant propagation by Tissue culture Part 1 & 2, Exegetics Ltd. Great Britain, U.K.
8. Maheshwari, P. and Rangaswamy, N.S. (Eds.) 1963. Plant, Tissue and organ culture – A symposium, Intern. Soc. Plant Morphologists, New Delhi.
9. Razdan, M.K. 1993. An introduction to plant tissue culture. Oxford and IBH Publishing Co. Pvt. Ltd. New Delhi.
10. Reinert, J.R., and Bajaj, Y.P.S. 1977. Applied and fundamental aspects of plant cell, tissue and organ culture. Springer-Verlag, Berlin.
11. Sen, S.K. and Giles, K.L. (Ed.) 1983. Plant cell culture in crop improvement. Plenum Press, New York.
12. Street, H.E. 1977. Plant tissue and cell culture. Academic Press, Berkeley, University of California. USA.
13. Thorpe, T.A. (Ed.) 1995. Embryogenesis in plants. Kluwer Academic Publishers, Netherlands.
14. Vasil, I.K. (Ed.). Cell culture and somatic cell genetics of plants, various volumes. Academic Publishers, Orlando.
15. Yeoman, M.M. 1985. Practical Cell culture technology. Blackwell Scientific Pub. London.

**Practicals**

1. In vitro morphogenetic studies on any one plant system (Seed culture, multiplication of shoots, rooting and hardening)
2. Isolation of explants, establishment, subculture and maintenance of callus.
3. Morphology of callus cells (callus smear preparation) and histological aspects (microtomy).
4. Embryogenesis in culture cells/tissues
5. Anther culture for haploid production.
6. Embryo culture.
7. Preparation of synthetic seeds.
8. Isolation of Protoplasts
9. Secondary Metabolite production by single cell culture

**Paper 15: Socioeconomic aspects and IPR (GMBT-403)****Course Objectives**

At the end of this course the students will be able to describe: 1. What is Indian patent law, WTO and how it is related with IPR. 2. Ethical and depository considerations in biotechnology, and What bioethics are and why those are needed in research,”

### **Course Content**

#### **UNIT – 1:**

Steps to preserve biodiversity. *In situ* and *Ex Situ* conservation - Gene banks, *In-situ* and *Ex situ* conservation. *Ex situ* conservation efforts at international level, *Ex-situ*, conservation by G-15 countries, Europe, India. Conservation efforts by private sectors, management of germplasm collection. Species conservations.

#### **UNIT – 2:**

**Biosafety and Societal Concern:** Public debate and concern on Genetically modified microorganisms, plants and animals, scientific analyses of the concern, Biosafety regulation and guidelines on developing and using the Genetically modified organisms, radiation safety.

#### **UNIT – 3:**

Intellectual property, Intellectual property rights (IPR) (Patents, trade secret, copy right, trade marks), Choice of intellectual property protection (IPP). IPR and plant genetic resources (PGR).

#### **UNIT – 4:**

Patenting of Biological Materials: International conventions. International cooperation obligations with patent applications, implications of patenting, current issues: Can live form be patented-? with special reference to Factor VIII, Erythropoietin, tissue plasminogen, activator, hybridoma technology etc. Patenting of higher plants and animals: Transgenic organisms and isolated genes.

Patenting of genes and DNA sequences, plant breeder's rights and farmer's right.

#### **Recommended Books:**

1. Entrepreneurship: New Venture Creation : David H. Holt
2. Patterns of Entrepreneurship : Jack M. Kaplan
3. Entrepreneurship and Small Business Management: C.B. Gupta, S.S. Khanka, Sultan Chand & Sons.
4. Sateesh MK (2010) Bioethics and Biosafety, I. K. International Pvt Ltd.
5. Sree Krishna V (2007) Bioethics and Biosafety in Biotechnology, New age international publishers

#### **SEMINAR (GMBT-404)**

#### **PROJECT/DISSERTATION (GMBT-405)**

#### **EDUCATIONAL TOUR/FIELD WORK**