SRI VENKATESWARA UNIVERSITY: TIRUPATI

PROGRAMME: B.Sc. (Honours) in Biotechnology (Major)		
w.e.f AY 2023-2024		
Semester IV	Course Code:	
Course: IV-1: r DNA Technology		
Credits:3	3hrs	

I. Learning outcomes:

Students after successful completion of the course will be able to

- 1. Understand radiolabelling and DNA sequencing
- 2. Know about various types of restriction Enzymes
- 3. Gain knowledge about vectors
- 4. Familiarize with principle and applications in analysis of recombinants
- 5. Know about expression systems

II. Syllabus:

Theory

UNIT -1: Characterization of Nucleic Acids

DNA and RNA: Quantification, Radiolabelling of nucleic acids, labelling by primer extension, DNA sequencing: Maxam-Gilbert (Chemical) and Sanger- Nicolson (Dideoxy/ enzymatic) sequencing method, Pyrosequencing.

UNIT-2: Restriction Enzymes

Types and uses of restriction endonuclease, Restriction mapping. DNA modifying enzymes Nucleases, Polymerases, Phosphatases and ligases.

UNIT-3: Vectors

Plasmid vectors, Bacteriophage, expression vectors, other vectors, Construction of genomic and c-DNA libraries, Joining of DNA Fragments to vectors, cohesive and blunt end Ligation, adaptors, and linkers.

UNIT- 4: Principle and applications in analysis of recombinants

Principle of hybridization.Northern blotting, Southern blotting and Western blotting. Polymerase chain reaction, selection and screening of recombinants, Restriction fragments length polymorphism, RAPD, AFLP, MAP.

UNIT – 5: Expression systems

Methods of Transformation, Codon optimization, host engineering. Strategies of genedelivery, *in vitro* translation, expression in bacteria, yeast, expression in insects and mammalian cells. Applications of r DNA Technology

Practical syllabus: IV-1: r DNA Technology

Credits:1

2hrs/week

III. Skill outcomes:

On successful completion of the practical course, student shall be able to

- 1. develop skill on primer designing
- 2. learn plasmid DNA Isolation from E.coli
- 3. learnrestriction Digestion and analysis
- 4. learn the technique of competent cell preparation and bacterial transformation
- 5. learn to perform Agarose Gel Electrophoresis

IV. Practical syllabus:

- 1. Primer designing- A computer approach.
- 2. Plasmid DNA Isolation from *E.coli*
- 3. Restriction Digestion and analysis (web cutter).
- 4. Competent Cell preparation and bacterial transformation
- 5. DNA Ligation.
- 6. Bacterial transformation.
- 7. Agarose Gel Electrophoresis.
- 8. Quantifying DNA by zymogram (computer approach).
- 9. SDS PAGE.

V. References:

- 1. Principles of Gene manipulation (1994) Old R.N. and Primrose S.B.
- 2. From Genes to Clones (1987) Winnaeker E.L.
- 3. Recombinant DNA (1992) Watson J.D., Witreowski J., Gilman M. and Zooller M.
- 4. An Introduction to Genetic Engineering: Nicholl, D.S.T.
- 5. Molecular Biotechnology (1996) Pasternak
- 6. The Biochemistry of Nucleic acid(1996)Adam et al
- 7. Genetic Engineering (1998)Janke k. swtlow
- 8. Molecular cloning: Sambrook et al.

Theory

I. Learning outcomes:

Students after successful completion of the course will be able to

- 1. Understand concepts and applications of plant tissue culture
- 2. Learn about transgenic plants and molecular markers
- 3. Acquire knowledge regarding animal Tissue Culture Techniques
- 4. Apprehend the concepts and applications of IVF technology
- 5. Understand methods of transgenic Animal Production

II. Syllabus

UNIT -I: Basic aspects of Plant Tissue Culture

Introduction to plant tissue culture: Preparatory techniques - cleaning, sterilization, sterile handling tissue culture lab requirements. Totipotency, Media & Composition, Sterilization techniques, Establishment of cultures: Callus and suspension cultures. Organogenesis and plant regeneration.Somatic embryogenesis.

UNIT-II: Applications of Plant Tissue Culture

Haploid plants Production, Virus Free Plants Production, Micropropagation, Protoplast culture, Somatic hybridization, and, Plant Secondary Metabolites- production & importance. Germplasm preservation.

UNIT-3: Animal Tissue Culture Techniques

Animal cell culture: Culture media and types, Culture of mammalian cells, Primary culture, Secondary cultures, cell lines. Stem Cells: Types, Culture and Applications. Cell Viability and Toxicity tests, Cryopreservation, Gene Transfer Methods.

UNIT- 4: Assisted Reproductive Technology

Human Embryo Development, Overview of IVF: Artificial insemination, Ovum retrieval, ICSI, ZIFT, Embryo Transfer, GIFT, Surrogacy, Pre Implantation Genetic Diagnosis, Advantages and Limitations, Ethical issues.

UNIT – 5: Transgenic Animals & Gene Therapy

Methods of Transgenic Animal Production & Examples.Recombinant production of Insulin, Somatostatin&Vaccines. Gene Therapy and it types. Applications of Biotechnology in human and animal health care.

Practical

Credits:1

2hrs/week

III. Skill outcomes:

On successful completion of the practical course, student shall be able to:

- 1. Perform Plant cell culture media preparation
- 2. Learn the skill to perform callus Culture, micropropagation
- 3. Prepare synthetic Seeds
- 4. Will do RAPD and RFLP analysis
- 5. Perform cell viability tests, cell Counting

IV. Practical syllabus:

- 1. MS Media preparation
- 2. Callus Culture
- 3. Micropropagation
- 4. Protoplast Isolation
- 5. Preparation of Synthetic Seeds
- 6. Animal cell culture Media preparation
- 7. Cell Viability tests
- 8. Cell Counting
- 9. Culture of Chick Embryo fibroblast

V. References:

- 1. Introduction to Plant tissue Culture. MK Razdan, 2003
- 2. Plant Tissue Culture, Kalyan Kumar De, 199 M7, New Central Book Agency
- 3. Biotechnology by U. Satyanarayana
- 4. Plant cell, Tissue and Organ Culture: Applied & Fundamental Aspects: YSP Bajaj,
- A. Reinhard: 2001
- 5. A Text book of Biotechnology by RC Dubey, 2003
- 6. Elements of Biotechnology by PK Gupta (1994)Rastogi Publications
- 7. Daniel et.al; Stem Cell Biology, 200; CSHL press, New York
- 8. M MRanga, Animal Biotechnology: Agrobios (India) 2006

Semester IV Course: IV-3: MOLECULAR GENETICS

Course Code:

Theory

Credits:3

I. Learning outcomes:

Students will be taught Mendelian genetics, their principles and gene interaction.

- 1. They learn about chromosomal aberrations and structure of chromosomes
- 2. The student will gain a basic understanding on human genetics and hereditary.
- 3. The course teaches the students about genes at molecular level
- 4. They learn about DNA, RNA and their replication, mutations, DNA repair mechanism.
- 5. The course outcome is to train the students in understanding genetics and relate modern DNA technology for disease diagnostics and therapy

II. Syllabus

UNIT – I

Recapitulation of Mendelian Principles - Principles of Segregation; Laws of inheritance dominance, recessiveness, laws of segregation, Dihybrid and trihybrid ratios- laws of independent assortment-test cross and back cross. Incomplete dominance- eg: flower colour, chromosomal theory of inheritance. Extension to Mendel's laws- Multiple allelismeg. Coat colour in rabbits, eye colour in drosophila, ABO blood groups, Incompatability and pseudoallelism,

UNIT-II

XY-chromosomes- sex determination in drosophila, birds, man. X-linked inheritance, hemophilia, colour blindness, Y-linked inheritance- holandric genes. Mechanisms of sex determination – Simple Mechanisms, The balance concept of Sex determination.Mosaics and Gynandromorphs.Sex differentiation.Sex- influenced dominance; Sex-linked inheritance-Morgan's discovery of sex linkage in drosophila.Patterns of inheritance of Sex-linked genes.

Unit- III

Gene as a unit of expression: Modern concept of gene, co-linearity of gene and polypeptide, types of genes (constitutive, structural, regulatory, luxury, overlapping, split genes etc.,) Biology of plasmids : Types of plasmids, incompatibility grouping, control of copy number replication of Col E1 and F plasmid. Transposons: Transposable elements in prokaryotes and eukaryotes, types of bacterial transposons - insertional sequences, complex transposons, Mechanisms of transposition (Replicative and Non replicative), Transposable viruses and retroposons. Complex transposons-Tn10, Tn5, Tn9 and Tn3 as examples.and applications of transposition by simple and complex elements.

UNIT-IV

Mechanism of genetic transfer in bacteria: Transformation, Transduction, Conjugation. Mapping of bacterial chromosome. Genetic recombination in bacteria, models and mechanism, role of rec A proteins. Homologous Recombination, Holiday junction, gene targeting, gene disruption, FLP/FRT and Cre/Lox recombination, RecA and other recombinases.

Unit-V

Mutations and Mutagenesis: Types of mutations, molecular basis of mutations, mutagenic agents, and mechanism of mutagenesis. Transposon mutagenesis, Site directed mutagenesis. Evaluation of mutagens by Ames test. Genetics of Eukaryotes: Gene linkage and chromosome mapping, crossing over, three point cross, tetrad analysis, complementation. Organization of chromosomes, specialized chromosomes, chromosome abnormalities, qualitative inheritance, population genetics.Developmental genetics using Drosophila asmodel system.Somatic cell genetics. Viral genetics : Organisation of genome in Lambda, T4 phage, $\varphi x 174$ and M13.

III. Skill outcomes:

On successful completion of the practical course, student shall be able to:

- 1. Perform Lethality curve construction
- 2. Learn the skill to Screen and isolate streptomycin resistant mutant
- 3. Detect mutagens by Ame's test.
- 4. Will do Transfer of genes in bacteria by Transformation
- 5. Perform Transfer of Genes in bacteria by conjugation.

IV. Practical syllabus:

- 1. Screening and isolation of streptomycin resistant mutant by gradient plate technique.
- 2. Lethality curve construction
- 3. Induction of mutation in bacteria using UV light, chemical mutagens photoreactivation.
- 4. Detection of mutagens by Ame's test.
- 5. Transfer of genes in bacteria by Transformation
- 6. Transfer of Genes in bacteria by Transduction
- 7. Transfer of Genes in bacteria by conjugation.
- 8. Curing of plasmids from E.coli strains.

V. References

- 1. Benjamin Lewin. Gene VII. Oxford University Press, U.K., 2000
- 2. William H Elliott and D C.Elliolt., Biochemistry & amp; Molecular biology, Oxford
- 3. S.R. Maloy, J.E. Cronon, and D. Freifelder., Microbial Genetics Jones & amp; Bartlet 1996.
- 4. Streips U.N. and YasbinR.E, Modern Microbial Genetics. Wiley-liss, 1991.
- 5. Stent G.S.Calender R., Molecular Genetics. CBS publishers, 1986.
- 6. E.J.Gardner, D.P.Simmons, M.J.Snustad, Principles of Genetics, 8th ed.Johnwiley private Ltd., Singapore. 2003.
- 7. David Freifelder, Microbial Genetics. Narosa Publishing House, New Delhi 2000.
- 8. David freifelder and G.M.Malacinik, Essentials of molecular biology 1996,
- 9. David J Sheratt, Mobile Genetic Elements, Oxford University Press. 1995
- 10. J.W. Dale, "Molecular Genetics of Bacteria" Wiley & amp; Sons 1994.
- 11. D.L.G. Hartl, "Basic Genetics" Jones Publ., 1991.
- 12. M. P. Arora, "Fundamental of Genetics", Himalaya Publishing House, Mumbai, 2004.
- 13. C.B. Powar, "Genetics", Volume 1, Himalaya Publishing House, Mumbai, 2003.

MODEL QUESTION PAPER

Max. Marks: 75

Time: 3 hrs (Total: 5x5=25 Marks)

SECTION A (Answer any five questions. Each answer carries 5 marks (At least 1 question should be given from each Unit)

1.	
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SECTION B (Total: 5x10 = 50 Marks) (Answer any five questions. Each answer carries 10 marks (At least 1 question should be given from each Unit)

9.	А		
		or	
	В		
10.	А		
		or	
	В		
11.	А		
		or	
	В		
12.	А		
		or	
	В		
13.	А		
		or	
	В		