Dr. V. S. KRISHNA GOVERNMENT DEGREE AND PG COLLEGE (A), VSKP
DEPARTMENT OF BIOTECHNOLOGY
II B.Sc BIOTECHNOLOGY SYLLABUS
SEMESTER IV

Course IV – MOLECULAR BIOLOGY AND RECOMBINANT DNA TECHNOLOGY

(Total Hours of Teaching 60 @ 04hrs/week)
Credits - 04

Course Outcomes:

On successful completion of this course, the students will have the knowledge and skill to:

- Illustrate the structural levels of DNA and genome organization in prokaryotes and eukaryotes.
- Explain the concept of gene and gene architecture.
- Demonstrate the overview of the central dogma of life and various molecular events.
- Illustrate molecular events in DNA synthesis, RNA synthesis, and protein synthesis and the role of different enzymes.
- Explain the regulation of gene expression in prokaryotes using the operon concept.
- To illustrate the creative use of modern tools and techniques for manipulation and analysis of genomic sequences.
- To expose students to applications of recombinant DNA technology in biological research.
- To train students in strategizing research methodologies employing genetic engineering techniques.
- To provide sound knowledge on methodologies that allow students to innovatively apply these engineering techniques in basic and applied fields.

MODULE I: Genome Organization and DNA Replication 12 Hours

1.1 Watson and Crick model of DNA; Genome organization in prokaryotes and Eukaryotes.

1.2 Experiments to prove DNA as Genetic Material: Griffith's transformation, Avery-Mc.Cloid and Mc.Carthy's and Hershey & Chase experiments; RNA as Genetic material in Tobacco Mosiac Virus.

1.3 Enzymes of DNA Replication: DNA polymerase I, II and III, Helicases, Primase, Single Strand Binding Proteins, Topoisomerases, DNA ligases.

1.4 Semiconservative replication; Replication origins, Replication mechanism: Initiation, Elongation and Termination.

MODULE II: Gene Expression and regulation 12 Hours

2.1 Transcription: Features; Structure and functions of RNA polymerases, Prokaryotic and Eukaryotic Promoters.

2.2. Transcription Mechanism in Prokaryotes and Eukaryotes: Initiation, Elongation and Termination.

2.3. Regulation of Gene Expression: Operon concept, Lac-operon, Trp-operon.

2.4. Post transcriptional modifications: capping, poly-adenylation

MODULE III: Genetic code and Protein Synthesis 12 Hours

3.1.Genetic code: Features of Genetic code; Coding dictionary; Codon-anticodon interaction- Wobble hypothesis.

3.2. Features and Structure of tRNA and mRNA and rRNA

3.3. Translation: prokaryotic and eukaryotic translation: attachment of amino acid to tRNA, initiation, elongation and termination mechanism.

3.4. Post translational modifications: Glycosylation, Acetylation and Ubiquitination.

MODULE IV: Recombinant DNA technology 12 Hours

4.1. Restriction and Modification: Classification of restriction endonucleases. Enzymes used in molecular cloning; Polymerases, ligases, phosphatases, kinases and nucleases

4.2. Blotting techniques: Southern, Northern and Western blotting techniques.

4.3. Cutting and joining DNA**:** cohesive end ligation, methods of blunt end ligation.

4.4. Transfection and transformation. Selection of transformed cells. Screening methods (Genetic marker and blue white screening

MODULE V: Cloning Vectors and Gene Sequencing 12 Hours

5.1. Cloning vectors: Plasmids, cosmid, PBR322, Bacteriophage, phagemid and shuttle vectors; Vectors of cDNA library construction: lambda phage vectors, cosmids, BAC & YAC; construction of cDNA library.

5.2. Gene transfer techniques: microinjection, micro-projectile bombardment-gene gun method, Electroporation and Agrobacterium mediated transformation

5.3. Methods of gene sequencing – Maxam - Gilberts and Sanger's dideoxy chain termination methods; Polymerase chain reaction technique.

5.4. Applications of rDNA technology: agriculture, industrial, pharmaceutical and medicine.

PRACTICALS
SEMESTER -IV
Course IV – MOLECULAR BIOLOGY AND RECOMBINANT DNA TECHNOLOGY
(Total hours of Laboratory Exercises – 30 hours @ 03 hrs/week)
Credits - 02

Course Outcomes:

On successful completion of this course, the students will have the knowledge and skill to:

- Demonstrate the absorption spectrum of DNA and RNA using Spectrophotometer.
- Estimate DNA and RNA quantitatively along with graphical representation.
- Isolate plasmid and genomic DNA from bacteria.
- Demonstrate the separation of DNA
- Restriction digestion, ligation and electrophoretic separation of DNA
- Perform nuclease enzyme activities
- Isolate plasmid DNA from bacteria
- Solve genetic engineering problems

Practical Syllabus

1. Quantitative estimation of DNA
2. Quantitative estimation of RNA
3. Isolation of plasmid DNA from bacteria
4. Isolation of genomic DNA from bacteria
5. Separation of DNA by Agarose gel Electrophoresis
6. Problems in Genetic engineering.
7. Transformation in Bacteria.
8. Restriction digestion of DNA and its electrophoretic separation.
9. Ligation of DNA molecules and their testing using electrophoresis.