

MMB - C201
MOLECULAR BIOLOGY AND RECOMBINANT DNA TECHNOLOGY

L T Credit
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Learning objectives:

- To make student understand about the structure and function of biologically important molecules.
- To know the historical background of DNA structure and its role as genetic material.
- To know about DNA, RNA and the molecular events that govern cell functions.
- Become familiar with different tools and techniques used in genetic engineering and recombinant DNA technology.
- To understand the applications of DNA modifying enzymes, cloning strategies, vector types, and screening of recombinants
- Students will know how genes express and regulate in prokaryotic cells.

Learning outcomes:

At the end of course student will be able to

- Explain why DNA is the genetic material of bacteria.
- Explain the application of genetic engineering techniques in basic and applied experimental biology.
- Amplify the DNA using PCR for the diagnosis and DNA fingerprinting.
- Describe how protein synthesis occur in procaryotic cell and enzyme involved in it.

UNIT - I

Nature of Nucleic acids- Nucleic acids as genetic material (evidences from bacteria, bacteriophages, bacterial conjugation, RNA viruses); DNA structure- historical aspects and current concepts, organization of DNA in eukaryotic cell, DNA torsion angles and sugar pucker; types of RNA- rRNA, mRNA (the 5' cap, non-coding region, initiation codon, coding region, termination codon, poly-adenylation); tRNA (structure of tRNA-clover leaf model).
(10 Lectures)

UNIT - II

DNA Replication, Damages and Repair Systems- Watson and Crick's model of DNA replication (experimental evidence), enzyme involved in DNA replication (DNA polymerase I, Pol II, Pol III, DNA ligase); mechanism of DNA replication; DNA damage and repair systems- types of damage (deamination, oxidative damage, alkylation, pyrimidine dimers); repair pathways- methylation -directed mismatch repair, very short patch repair, nucleotide excision repair, base excision repair, recombination repair, SOS repair.
(15 Lectures)

UNIT - III

Gene Expression and Regulation- Gene expression- RNA polymerase, site of transcription; transcription- chain initiation, chain elongation, chain termination; post-transcriptional processing of RNAs- methylation, polyadenylation and splicing of mRNA; translation- charging of tRNA, initiation of polypeptide synthesis, elongation of polypeptide chain, termination of polypeptide chain; gene regulation- negative regulation- *lac* operon of *E. coli* promoter, repressor and operator genes, structural gene.
(15 Lectures)

UNIT - IV

Cloning Enzymes and Vectors- Essential enzymes used in rDNA technology: nucleases, restriction endonucleases, alkaline phosphatases, DNA polymerase, terminal transferases, ligase, reverse transcriptase; restriction digestion, ligation; cloning vectors - plasmids cosmids, Ti plasmids, BAC vectors, YAC vectors; cloning strategies, gene libraries- cDNA and genomic libraries.
(10 Lectures)

UNIT - V

Blotting Methods and Gene Sequencing- PCR- working principle and applications; electrophoresis; blotting techniques- Southern blotting, Western blotting; Northern blotting, nucleic acid hybridization; gene sequencing methods: Maxam-Gilbert methods, Sanger & Nicolson method, automated gene sequencing.
(10 Lectures)

Suggested Reading

1. Bruce Alberts. Molecular Biology of the Cells, W.W. Norton and Company, ISBN: 9780815344643
2. Dubey, R.C. *Advanced Biotechnology*. S. Chand & Co. P Ltd, New Delhi, p. 1161; ISBN: 81:219-4290-X.
3. Harvey, Lodish. *Molecular Cell Biology*, W.H. Freeman
4. Dubey, R.C. and Maheshwari, D.K. *Practical Microbiology*. 2nd ed., S. Chand & Co. P Ltd, New Delhi, p. 413. ISBN: 81:219-2559-2

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