

DSE- 8 Recombinant DNA Technology

MM : 100

Time : 3 hrs

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Total Hours: 60

Sessional : 30

ESE : 70

Pass Marks : 40

Learning objectives:

- To make students clear about the structure and function of biologically important molecules.
- To know the historical background of DNA structure and its role as genetic material.
- To 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 gene expresses and regulates in prokaryotic cells.

Learning outcomes:

At the end of course students will be able to

- Explain why DNA is the genetic material.
- 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 prokaryotic cell and enzyme involved in it.

UNIT-I**(16 Lectures)****Introduction to Genetic Engineering**

Milestones in genetic engineering and biotechnology; Molecular Cloning- Tools and Strategies-Cloning Tools; Restriction modification systems: Types I, II and III. Mode of action, nomenclature, applications of Type II restriction enzymes in genetic engineering DNA modifying enzymes and their applications: DNA polymerases. Terminal deoxynucleotidyl transferase, kinases and phosphatases, and DNA ligases Cloning Vectors: Definition and Properties of Plasmid vectors: pBR, Cosmids, Expression vectors.

UNIT- II**(14 Lectures)****Methods in Molecular Cloning**

Transformation of DNA: chemical method, electroporation, Gene delivery: Microinjection, electroporation, biolistic method (gene gun), liposome and viral mediated delivery, *Agrobacterium* - mediated delivery DNA, RNA and Protein analysis: Agarose gel electrophoresis, Southern - and Northern – blotting techniques, DNA Western blotting.

UNIT- III**(09 Lectures)****DNA Amplification and DNA Sequencing PCR**

Basics of PCR, Real-Time PCR, Sanger's method of DNA Sequencing: traditional and automated sequencing.

UNIT- IV**(09 Lectures)****Construction and Screening of Genomic and cDNA Libraries**

Genomic and cDNA libraries: Preparation and uses, Screening of libraries: Colony hybridization and colony PCR.

UNIT - V**(12 Lectures)****Applications of Recombinant DNA Technology**

Products of recombinant DNA technology: Products of human therapeutic interest - insulin, hGH, antisense molecules. Bt transgenic - cotton, brinjal, Gene therapy, recombinant vaccines, protein engineering and site directed mutagenesis.

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Suggested Reading

1. Bruce Alberts. Molecular Biology of the Cells, W.W. Norton and Company, ISBN: 9780815344643
2. Harvey, Lodish. Molecular Cell Biology, W.H.Freeman.
3. Sambrook J, Fritsch E. F. and Maniatis (1989) Molecular cloning, vol. I, II, III, II nd edition, Cold spring harbor laboratory press, New York.
4. DNA Cloning : A practical approach D.M. Glover and D.B. Hames, RL Press, Oxford, 1995
5. Molecular and cellular methods in Biology and Medicine, P.B. Kaufman, W. Wu , D. Kim and L.J. Cseke, CRC Press Florida 1995.
6. Methods in Enzymology Guide to Molecular Cloning Techniques, Vol. 152 S.L. Berger and A. R. Kimmel, Academic Press Inc, San Diego, 1996
7. Methods in Enzymology Gene Expression Technology, Vol. 185D. V. Goedel, Academic Press Inc, San Diego, 1990
8. DNA Science: A First Course in Recombinant Technology, D. A. Mickloss and G. A Freyer, Cold Spring Harbor Laboratory Press, New York, 1990
9. Molecular Biotechnology, 2nd Ed. S. B. Primrose, Blackwell Scientific publishers, Oxford, 1994
10. Milestones in Biotechnology, Classic Papers on Genetic Engineering, J. A. Davis and W. S. Reznikoff, Butterworth-Heinemann Boston 1992
11. Route Maps in Gene Technology, M. R. Walker, and R. Rapley, Blakwell Science, Oxford, 1997
12. Genetic Engineering : An Introduction to Gene Analysis and Exploitation in Eukaryotes, S. M. Kingsman, Blackwell Scientific Publications, Oxford, 1998
13. An Introduction to Genetic Engineering, 3rd Edition. Desmond S. T. Nicholl, Cambridge University press, 2008.
14. Gene Cloning and Manipulation, 2nd Ed. Cristopher Howe, Cambridge University Press, 2007.

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