Course: Discipline Specific Core

Semester ____	6
Paper Number	HMBCR6142T/P
Paper Title	**Recombinant DNA technology**
No. of Credits	6 (Th:4, Pr:2)
Theory/Composite	Composite
No. of periods assigned	Th: 4 Pr: 3
Course description/objective	1. To understand the different methods employed in producing Recombinant DNA molecules. 2. Different applications of RDT.
Reading/Reference Lists	1. Brown TA. (2010). Gene Cloning and DNA Analysis. 6th edition. Blackwell Publishing, Oxford, U.K. 2. Clark DP and Pazdernik NJ. (2009). Biotechnology: Applying the Genetic Revolution. Elsevier Academic Press, USA 3. Primrose SB and Twyman RM. (2006). Principles of Gene Manipulation and Genomics, 7th edition. Blackwell Publishing, Oxford, U.K. 4. Sambrook J and Russell D. (2001). Molecular Cloning-A Laboratory Manual. 3rd edition. Cold Spring Harbor Laboratory Press 5. Wiley JM, Sherwood LM and Woolverton CJ. (2008). Prescott, Harley and Klein's Microbiology. McGraw Hill Higher Education 6. Brown TA. (2007). Genomes-3. Garland Science Publishers 7. Primrose SB and Twyman RM. (2008). Genomics: Applications in human biology. Blackwell Publishing, Oxford, U.K.
Evaluation	CIA: 20 End-Sem: 80 (Th:50 and Pr:30) Question paper format of Th paper (Mod 1: 30 Mod 2: 20 Marks) Module 1 with 30 marks: Objective questions 6 marks (6 questions out of 8) 3 questions of 8 marks each (3 questions out of 4) Module 2 with 20 marks: Objective questions 4 marks (4 questions out of 6) 2 questions of 8 marks each (2 questions out of 3)

C-14: RECOMBINANT DNA TECHNOLOGY (THEORY)
SEMESTER –VI

HMBCR6142T

TOTAL HOURS: 52 **CREDITS: 4**

Module 1 **Marks 30**

Unit 1 Molecular Cloning- Tools and Strategies **No. of Hours: 20**
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
Plasmid vectors: pBR and pUC series
Bacteriophage lambda and M13 based vectors
Cosmids, BACs, YACs
Use of linkers and adaptors
Expression vectors: *E.coli* lac and T7 promoter-based vectors, yeast YIp, YEp and YCp vectors, Baculovirus based vectors, mammalian SV40-based expression vectors

Unit 2 DNA Amplification and DNA sequencing **No. of Hours: 10**
PCR: Basics of PCR, RT-PCR, Real-Time PCR
Sanger's method of DNA Sequencing: traditional and automated sequencing
Primer walking and shotgun sequencing

Module 2 **Marks 20**

Unit 3 Methods in Molecular Cloning **No. of Hours: 12**
Transformation of DNA: Chemical method, Electroporation,
Gene delivery: Microinjection, electroporation, biolistic method (gene gun), liposome and viralmediated delivery, *Agrobacterium* - mediated delivery
DNA, RNA and Protein analysis: Agarose gel electrophoresis, Southern - and Northern - blotting techniques, dot blot, DNA microarray analysis, SDS-PAGE and Western blotting.

Unit 4 Construction and Screening of Genomic and cDNA libraries No. of Hours: 5

Genomic and cDNA libraries: Preparation and uses, Screening of libraries: Colony hybridization and
colony PCR, Chromosome walking and chromosome jumping

Unit 5 Applications of Recombinant DNA Technology No. of Hours: 5

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 mutagensis

C-14: RECOMBINANT DNA TECHNOLOGY (PRACTICAL)
HMBCR6142P

TOTAL HOURS: 39 CREDITS: 2

1. Preparation of competent cells for transformation
2. Demonstration of Bacterial Transformation and calculation of transformation efficiency.
3. Digestion of DNA using restriction enzymes and analysis by agarose gel electrophoresis
4. Ligation of DNA fragments
5. Cloning of DNA insert and Blue white screening of recombinants.
6. Interpretation of sequencing gel electropherograms
7. Designing of primers for DNA amplification
8. Amplification of DNA by PCR
9. Demonstration of Southern blotting