BIRLA INSTITUTE OF TECHNOLOGY & SCIENCE, PILANI, HYDERABAD CAMPUS INSTRUCTION DIVISION, FIRST SEMESTER 2016-2017 Course Handout (Part II)

Date: 01.08.2016

In addition to part I (General Handout for all courses appended to the time table) this portion gives further specific details regarding the course.

Course No. : BIO F311

Course Title : Recombinant DNA Technology

Instructor : S. Swaminathan

- **1. Course Description:** Recombinant DNA technology is an interdisciplinary field encompassing biochemistry, microbiology, immunology, molecular biology, genetic engineering, cell biology and chemical engineering. This course will deal with theoretical aspects underlying the practice of recombinant DNA technology. It will focus mainly on the tools and techniques available to create and manipulate chimeric DNA molecules.
- **2. Scope and Objective:** This course intends to provide interested students an opportunity to gain basic theoretical understanding of recombinant technology through lectures, interactive tutorials, and quizzes. It aims to introduce the students to the versatile tools and techniques of genetic engineering and recombinant DNA technology that can be applied to virtually any research question that involves a molecular approach. The specific learning objectives are listed in the table below.
- **3. Text Book (T1):** Gene Cloning & Manipulation, by Christopher Howe, 2nd Edition (First South Asia Edition, 2016), Cambridge University Press
- **4. Reference Book (R1):** S B Primrose and R.M. Twyman Principles of Gene Manipulation and Genomics. 7th Edition, 2006; Blackwell Publishing;

Reference Book (R2: Sambrook and Russell. Molecular Cloning: A laboratory Manual. Vols 1-3, 2001; CSH Press.

Course Plan

Lect.	Learning objective	Topics to be covered	Ref.
1-2	Introductory concepts	Over view of rDNA technology	R1, Chapter 1;
3	Chemical synthesis of DNA	Review of nucleic acid chemistry; Phosphodiester chemistry-based synthesis	Lecture notes
4-5	Basic tools & techniques	Cutting DNA; restriction endonucleases; other DNA modifying enzymes; joining DNA; ligases; Plasmid transformation, purification and electrophoresis; blotting techniques.	T1, Chapter 1 R1, Chapter 2
6-8	Polymerase Chain Reaction	Principle of PCR; primer design; melting temperature; applications of PCR; modifications of PCR: inverse PCR; RNA-PCR; real-time PCR.	T1, Chapter 2 R1, Chapter 2
9-11	DNA sequencing	Principle of sequencing; Sanger sequencing; cycle sequencing; pyrosequencing; analysing DNA sequence data	Lecture notes R1, Chapter 7
12-17	Plasmid vectors	Plasmid biology; plasmid replicons & copy number; examples of common plasmids; selection markers; cloning and expression vectors; expression modules;	T1, Chapter 3 R1, Chapter 4
18-21	Cloning: making recombinant DNA molecules	Basic cloning; sticky and blunt ends; ligation; linkers & adaptors; homopolymer tailing; TA cloning; cloning PCR products	T1, Chapter 3 R1, Chapter 3;
22-26	Making libraries	Construction of genomic & cDNA libraries; cloning full-length cDNA; directional cloning; 5' and 3' RACE;	T1, Chapter 5 R1, Chapter 6
27-29	Library screening	Library screening; screening by hybridization; PCR screening; expression screening; functional screening; Southwestern and Northwestern screening; screening for other functions	T1, Chapter 6 R1, Chapter 6
30-32	Modification & mutagenesis; protein engineering	Restriction enzyme-based and oligonucleotide-directed mutagenesis; The single primer method; PCR-based mutagenesis; gene inactivation techniques; creating chimeric proteins: gene shuffling; ITCHY and Thio-ITCHY methods;	T1, Chapter 7 R1, Chapter 8

33-34	Expression of cloned DNA	Expression of RNA and protein using cloned DNA; Expression strategies; purification of recombinant proteins; reporter genes and tags	T1, Chapter 8 R1, Chapter 6
35-36	Alternate vectors for <i>E. coli</i>	Bacteriophage and cosmid vectors; molecular aspects of lambda and M13 life cycle relevant to vector design; BACs and PACs	T1, Chapter 4 R1, Chapters 4, 5
37-38	Cloning in bacteria other than <i>E. coli</i>	Gram stain; self-transmissible gene transfer; bacterial transposons; IncP, IncQ and IncW plasmids for cloning in Gram negative (other than <i>E. coli</i>); cloning in Gram positive bacteria; mutigene assembly using <i>B. subtilis</i>	Lecture notes R1, Chapter 10
39-42	Gene transfer into animal cells	Different methods of DNA transfection (physical and chemical); transient and stable transfection; selection markers (endogenous; dominant, amplifiable markers); viral vectors and introduction to their design	T1, Chapter 9 R1, Chapter 12

5. Evaluation Scheme:

Evaluation Component	Duration	Weightage (%)	Date & Time	Remarks
Test 1	1 hour	20	10.9.16 (10-11 am)	Closed Book
Test 2	1 hour	20	22.10.16 (10-11 am)	Closed Book
Surprise quizzes*		20	Various	During lecture and/or tutorial classes
Comprehensive	3 hours	40	9.12.16 (AN)	Open Book (20%)**+Closed Book (20%)

^{*}Average of n-1 quizzes conducted will be taken (please see make-up policy below)

- 6. Chamber Consultation Hour: Will be announced in the Class.
- **7. Notices:** All notices, concerning the course will be displayed on CMS and/or the Biological Sciences Department Notice Board.
- **8. Grading policy:** Students missing one or more component of evaluation completely will be considered as having not cleared the course (NC grade).
- **9. Make-up policy:** For T-1, T-2 and Compre, make-up will be granted only if candidate is sick and hospitalized. <u>No make-up will be granted for surprise quizzes under any circumstances</u>.

INSTRUCTOR-IN-CHARGE BIO F311

^{**}Compre Open Book: This will be concept and understanding-based drawing mostly on actual topics discussed during both lecture and tutorial classes.