

# FIRST SEMESTER 2018-2019 Course Handout (Part II)

01.08.2019

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

Course No.: BIO F418

Course Title: GENETIC ENGINEERING TECHNIQUES

Instructor-in-Charge: K. NAGA MOHAN

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# 1. Course Description:

Experiments on the common molecular biology techniques used in gene manipulation in bacteria and plants; gene cloning procedure in bacteria – from isolation of plasmids to screening of recombinant clones; polymerase chain reaction (PCR) and its applications; gene and protein expression analysis; DNA sequencing; Agrobacterium-mediated gene transfer in plants; Use of software for molecular biology.

#### 2. Scope and Objectives of the Course:

This course aims to give the student hands-on experience of the essential techniques used in the molecular biology laboratory, with specific emphasis on DNA manipulation. The student would learn the theoretical bases behind the various experiments that he/she performs.

## 3. Textbook and Reference Books:

**Textbook (T)**. Metzenberg, Stan. Working with DNA. Oxford: Taylor and Francis, 2007. **Reference Book (R)**. Sambrook J., MacCallum P. and Russell D. Molecular Cloning: A Laboratory Manual (3rd edition, three-book set). New York, USA: CSHL Press, 2001.

## 3. List of Experiments:

(Note: Experiments may not necessarily be done in the order listed below)

- 1. Solutions and buffers
- 2. Preparation of *E.coli* competent cells-I
- 3. Preparation of *E.coli* competent cells-II
- 4. Determination of transformation efficiency using competent cells



- 5. Understanding a cloning experiment
- 6. Amplification of cDNAs from cloned plasmids
- 7. Preparation of insert DNAs for cloning
- 8. Isolation of plasmid DNA
- 9. Midipreps of plasmid DNA
- 10. Preparation of plasmid DNA for cloning
- 11. Setting up ligations and transformation of competent cells
- 12. Screening colonies for identification of recombinant plasmids
- 13. Sequence verification of the recombinant clones.
- 14. Scaling up the recombinant plasmid clones and their purification.

# 4. Lecture Plan:

Lect.#	Learning objective(s)	Topics to be covered	Chapter in the Text Book
1	Getting oriented to the course	Introduction to the course; mode of operation in the lab; how to maintain the lab record notebook.  Overview of the gene cloning procedure	1-3 (T) 1 (R)
2	Knowing about the bacterial host <i>E. coli</i>	<i>E. coli</i> and its versatility; knowing genotypes of strains; transformation procedures for <i>E. coli</i>	4 (T), 5 (R)
3	To know about prokaryotic vectors for gene cloning	<ul><li>(i) Plasmids – types, characteristics of an ideal cloning vector</li><li>(ii) Other cloning vectors - cosmids, phage vectors, etc.</li></ul>	4 (T) 5 (R)
4	Isolating and analyzing DNA	<ul><li>(i) Purification of plasmid DNA</li><li>(ii)Purification of genomic DNA from bacteria and phages</li><li>(iii) Quantitation and electrophoresis of DNA</li></ul>	4 (T) 3 (T) 2 (T)
5	Learn about restriction enzymes	Restriction enzymes and DNA digestion; restriction mapping	5 (T)
6	Other enzymes for DNA manipulation	DNA ligase, polymerase, phosphatase, kinase, topoisomerase.	6 (T), 4 (R)
7	Procedures for selecting the right clone	Direct selection; selection from gene libraries	4 (T), 8 (R)
8	Polymerase Chain Reaction	How to perform a PCR reaction in the lab; Primer design; post-PCR analysis – results vs. artifacts	7 (T, R) 6 (R)
9-10	Common techniques used in DNA and gene analysis	Southern, Northern, RT-PCR, SAGE, mutagenesis, etc.	8 (T) 3,6,8 (R)
11	Protein expression	Expression hosts, vectors and techniques (SDS-PAGE and Western blotting)	4 (T) 6,10 (R)



12	Some applications of genetic engineering	Sequencing and analyzing genomes, medical and forensic applications	3,9,11 (R)
13	Putting it all together; Feedback session	Use of various methods of gene cloning in research and biotechnology	8 (T)

#### 5. Evaluation scheme:

Component	Duration	Marks	%	Date and Time	Venue	Nature of the
						Component
Mid-sem	90 min	40	20	05.10.19 (1.30 – 3.00 PM)		Closed book
<b>Laboratory Evaluation</b>	-	100		TBA		TOTAL
a) Lab quizzes		40	20			Announced
						at a later date
b) Experiment-based		60	30			Announced
evaluation and viva						at a later date
Comprehensive exam	120 min	60	30	14.12.19 (FN)		Closed book

#### Notes:

(*i*) For Observation component: Every student would be assessed on the following criteria during the regular lab sessions: how successful and efficient is the student in doing the assigned experimental tasks, scientific integrity, punctuality to the lab, maintenance of lab decorum and ability to work in a group. Besides the regular assessment, pre-announced laboratory assignments could also be given.

## 6. Attendance Policy:

It is expected that the student attends every laboratory session and theory class. Individual students may be assigned specific tasks, forming part of the planned experiment, to be done before or during the lab hours, the completion of which may be required for the entire class group. If failure to complete the task due to absence is anticipated, it is the student's responsibility to inform the instructor prior to the scheduled laboratory.

#### 7. Grading Policy:

Award of grades would be guided in general by the histogram of marks. Decision for borderline cases would be based on the individual's sincerity, attendance in classes and the instructor's assessment of the student's capability.

- 8. Office Consultation Hour: To be announced in the class.
- **9. Make-up Policy:** Clause 4.07 of BITS *Academic Regulations* booklet should be consulted. Make-up can be requested only for the two class tests.

#### 10. Notices:

All course announcements shall be displayed in CMS and/or in the Biological Sciences departmental notice board only.

11. **Academic Honesty and Integrity Policy**: Academic honesty and integrity are to be maintained by all the students throughout the semester and no type of academic dishonesty is acceptable.

INSTRUCTOR-IN-CHARGE BIO F418

