Birla Institute of Technology & Science, Pilani
Hyderabad Campus

FIRST SEMESTER 2022-2023
Course Handout (Part II)

Date: 24.08.2022

Course No. : **BIO G512**
Course Title : **Molecular Mechanism Of Gene Expression**
Instructor-in-Charge : **K. NAGA MOHAN**

Instructors : **Anuhya Anne and Sushma S Kumar**

1. Course Description:

Prokaryotic and eukaryotic genomes and their topology: DNA - protein interactions; RNA transcription and transcriptional control; DNA replication; transcription in yeast; RNA processing; translation; mechanism of gene expression in pro and eukaryotes..

2. Scope & Objective:

The course is designed mainly to impart knowledge of how genomes are organized in bacteria, viruses and eukaryotes, chromatin structure and histone variants, replication and repair of genomic DNA, molecular tools used for studying gene expression, transcriptional mechanisms in prokaryotes and eukaryotes, epigenetic modifications influencing transcription in eukaryotes, post-transcriptional processing in eukaryotes, mRNA transport and regulation of mRNA levels in eukaryotes, translational mechanisms in prokaryotes and eukaryotes, regulation of gene expression in prokaryotes and eukaryotes. Through this course, the students would understand the genetic regulatory mechanisms in the context of various biological processes.

3. Text Book:

Lewin's Genes XII by Jocelyn E. Krebs, Elliott S. Goldstein, Stephen T. Kilpatrick, Jones and Bartlett Learning, 2017.

Reference Book:

1. Molecular Biology of Gene: Watson, Baker, Bell, Gann, Lavine&Losick (7th Ed).
2. Molecular Cell Biology: Harvey Lodish, Arnold Berk, S Lawrence Zipursky, Paul Matsudaira, David Baltimore, and James Darnell (2016) 8th edition, Macmillan learning
3. Research papers and reviews

4. Course Plan:

Lect No	Learning Objective	Topics to be covered	Text Book Chapter	Reference Book Chapters	
				RB 1	RB 2
1-3	**Understanding the basis of heredity, the nature and function of genetic material**	Evidence of DNA and RNA as genetic materials, genes encode polypeptides and RNAs, overview of transmission of genetic material and flow of genetic information	1	2	1
4-8	**Techniques used in**	Restriction endonucleases, Cloning	2	7	5

	studying gene expression	vectors, Detection of nucleic acids, DNA sequencing, PCR and RT-PCR, Blotting methods, DNA microarrays, Protein-DNA interactions, Chromatin immunoprecipitation, Gene knockouts, transgenics and Genome editing			
9-10	**Structures of DNA and RNA**	Overview of the DNA double helix, Alternative forms of DNA (A- and Z-DNA) their locations and functions, Structures of RNA and their functions.	-	4-5	4
4-7	**Organization of genes and genomes in prokaryotes and eukaryotes**	Prokaryotic genomes: Organization of genes in bacterial and viral genomes Eukaryotic genomes: Organization of genes and genomes in yeast and higher eukaryotes, C-value paradox, non-coding sequences and their importance	4-7	8	6
8-11		Eukaryotic chromatin: Nucleosomes-10nm and 30nm structures, histone variants and their functional role, organization into mitotic chromosomes and banding patterns, Centromere and telomeres.	8	8	6
12-18	**Maintenance of the genome**	DNA replication, recombination, repair, and transposition	9-16	9-12	4
19-27	**Transcriptional mechanisms**	Prokaryotes: Transcriptional initiation, elongation and termination. Eukaryotes: Transcriptional initiation, elongation, termination, RNA splicing and processing, mRNA stability, catalytic RNA	17-21	13-14	7-8
28-30	**Translation and genetic code**	Translational mechanisms in prokaryotes and eukaryotes, nature of the genetic code	22-23	15-16	4
31-35	**Regulation of gene expression**	Prokaryotes: Regulation of *lac* operon, regulation of *trp*operon, regulation of lytic and lysogenic phases in bacteriophages	24-25	18	7-8
36-40		Eukaryotes: mechanisms transcriptional activation, epigenetic regulation and regulatory RNA, Gene regulation during development, Large-scale gene silencing	26-30 Class Notes	19-21	7-8

Laboratory plan:
The main objective of the experiments planned is to relate the understanding of genome composition and epigenetic dynamics associated with gene expression. To develop analytical skills, the students are first asked to read about the subject matter, discuss in the class on the experiment to be conducted, its purpose and anticipated results. The students are then asked to write their own observations and inferences in own language as pdf files and not through mutual discussions and copying. Plagiarism check will be made and accordingly marks are awarded.

S. No.	List of experiments
Part I: Understanding the dinucleotide composition in the context of vertebrate genome evolution.	
1	Reagent and plasticware preparation
2	Isolation of genomic DNA from mammalian cells
3	Checking the quality of genomic DNA and its quantification
4	Use of restriction enzymes in studying the dinucleotide (CG, GC and TA) abundance
5	Visualization of repeat sequences by restriction enzyme digestion
Part II: Understanding the levels of DNA methylation in repeat elements, an imprinted gene and an autosomal promoter CpG island.	
6	Estimation of DNA methylation in genomic DNAs by digestion with methylation-sensitive, insensitive, and specific enzymes.
7	Designing primers for methylation analysis using bisulfite-treated DNA.
8	Estimation of DNA methylation levels in the IAP repeat sequence, an autosomal and an imprinted gene (*Snrpn*).
9	Understanding the experimental regulatory systems to control gene expression in mammals: Using the *tet* off system to turn off *Dnmt1* transcription using doxycycline and measuring the resultant *Dnmt1* transcript levels in the treated cells.
10	Comparing the effects of loss of *Dnmt1* transcripts on DNA methylation levels in IAP elements, autosomal and imprinted genes.
11	Optional experiment: Measurement of transcript levels of the target sequences listed for experiment 8.

5. Evaluation Scheme:

No	Evaluation Component	Duration	Date and Time	Weightage (%)	Remarks
1	Mid Sem	1.5 hours	01/11 1.30 - 3.00PM	25%	CB
2.	Practical components: Lab Notes and the quality of the experimental results obtained.	Variable	-	25%	OB
3.	Explanation of Results (Observation and Inference) and quiz	Variable	-	15%	OB

	based on the experiments conducted.				
4.	Comprehensive Examination	3 hours	21/12 FN	35%	CB

CB: Closed Book examination OB: Open Book examination

6. Chamber Consultation Hour: To be announced in the class

7. Notices: Notices will be displayed on the Course Management System (CMS)

8. Make-up Policy: Make up will be granted only for valid reasons with prior permission from the Instructor In-charge.

9. 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 G512