

## Syllabus for Dr. Freeman's Section of MCB502

Lecture 1: DNA structure; RNA Polymerase properties; Prokaryotic RNA Polymerase subunits;  $\sigma$  factor 70

**Techniques:** Filter binding assay; Methylation-S1 nuclease; Electro-Mobility Shift Assay (EMSA); DNA Radio-labeling; UV Crosslinking; DNA footprinting; Förster resonance energy transfer (FRET); Run-Off transcription assay

Lecture 2: Transcription termination;  $\sigma$  factor cascades; Transcription factors; Lac Operon

**Techniques:** Promoter bashing; Linker-scanning mutagenesis; Genetic screening

Lecture 3: DNA bending; Activation mechanisms; Promoter classes; Combinatorial Regulation

**Techniques:** Site-directed mutagenesis; DNA bending EMSA; Chromatin Immunoprecipitation (ChIP) assay; Genetic suppressor screens

Lecture 4: Proximal regulation by DNA bending; Distal regulation by DNA looping; Kinase signaling; Bacterial histone-like proteins; Packaging of prokaryotic genomes

**Techniques:** Plasmid concatemers; Topoisomerase assay; Fluorescent proteins; STORM imaging; 3C assay

Lecture 5: Lambda genetic switch

**Techniques:** Phage genetic screen; Northern blots; Real time RT-PCR; DNA Microarrays; RNA-Seq

Lecture 6: **\*\*First Exam in class\*\***

Lecture 7: Eukaryotic RNA Polymerase; General transcription factors; Basal promoter elements; CTD phosphorylation; Post-initiation events

**Techniques:** Epitope-tag pull-down assay; Helicase assay

Lecture 8: DNA packaging; Chromatin and transcription; Position effect variegation; Histone code; Boundary elements; CpG islands as markers of cancer

**Techniques:** Micrococcal nuclease assay; DNase I assay; Bisulfite PCR

Lecture 9: Phosphate signaling and the *PHO5* chromatin model; Lysine acetyltransferases; Chromatin remodelers; Histone chaperones

**Techniques:** In vitro chromatin templates

Lecture 10: Signaling pathways; Agonists & Antagonists; Transcription factor cascades; Intracellular hormone receptors; Combinatorial regulation; Response elements; Synergy

**Techniques:** Protein domain mapping; DNase-Seq

Lecture 11: Transcriptional coactivators; Post-translational modifications (PTMs)

**Techniques:** Two-hybrid; GST pull-down

Lecture 12: Ordered and cyclical transcription pathways; Utilization of PTMs

**Techniques:** Nuclear run-on assay

Lecture 13: Dynamic protein promoter complexes in vivo; Nuclear roles of molecular chaperones; Self-assembly vs. self-organization

**Techniques:** FISH; GFP; FRAP, FLIP, FLIM, Photoactivation, Protein Microarrays

Lecture 14: Genome organization; 3D genomes; Nuclear organization during cell cycle; Pioneering factors

**Techniques:** Chromosome conformation capture (3C) based assays

Lecture 15: **\*\*Second In-class Exam\*\***