## M. Sc II

## **SEMESTER III**

## BCH3T009: ADVANCED MOLECULAR BIOLOGY

### Unit I: Regulation of eukaryotic gene expression at transcriptional level

Overview of transcription by RNA Polymerases I, II, and III

Anatomy of a protein-coding gene

Basal transcription by RNA polymerase II: Subunits of Pol II; general transcription factors; Activators, How the initiation complex is assembled, How initiation occurs.

Speeding up the process: Enhancers, TAF's and how they work

Regulated transcription: transcription factors: Zinc-fingers (Sp1; the first such factor identified) Leucine zippers, Basic helix loop helix, Homeodomains, DNA binding domains, Activating domains

RNA Elongation: HIV TAT/TAR

RNA polymerase III and regulation of 5S rRNA

### Unit II: Regulation of eukaryotic gene expression at translational level

An overview and the elongation cycle, the ribosome as a molecular machine

Initiation and Regulation: Over-view of initiation of translation

Regulation of Translation: Global regulation through eIF2 and eIF4E/eIF4E-BP. Specific regulation through 5' UTRs using RNA structure e.g. ODC. Specific regulation through 5' UTR/protein interactions e.g. ferritin in eukaryotes and ribosomal proteins in prokaryotes. Specific regulation through 3' UTRs e.g. 15-LOX

### **Unit III: Regulatory RNAs**

Historical background, RNA interference as regulatory mechanism in eukaryotes Slicer and dicer, synthesis and function of RNAi molecules in plants, chromatin remodeling in human disease and diagnosis

## **Unit IV: Epigenetics**

Background, chromosomal inheritance taking fission yeast as an example, DNA methyltransferases, DNA methylation maintenance, histone modification and regulation of chromatin structure, bivalent histones, DNA demethylation, histone demethylation

- 1. Molecular Biology of the Cell: Alberts 5<sup>th</sup> Edition 2007 NCBI Publication
- 2. Principles of Biochemistry: Lehninger WH Freeman
- 3. Biochemistry of Signal Transduction and Regulation Gerhard Krauss Wiley VCH 3<sup>rd</sup> Revised Edition
- 4. Molecular Cell Biology: Lodish 6<sup>th</sup> Edition, WH Freeman & Company
- 5. The cell: Cooper 2<sup>nd</sup> Edition ASM Press
- 6. Genes IX: Benjamin Lewin Published by Pearson Prentice Hall
- 7. Cell and Molecular Biology: Gerald Karp
- 8. Molecular Biology: Robert Weaver 1<sup>st</sup> Edition, WCB McGraw-Hill
- 9. Molecular Biology of the Gene: Watson 6<sup>th</sup> Edition, Pearson Publication
- 10. Gene Regulation: A Eukaryotic Perspective: David Latchman 5 illustrated , Taylor & Francis, 2005

# **BCH3T010: BIOTECHNOLOGY**

### Unit I: Gene control systems in bacteria and bacteriophage lambda:

Mechanism of induction and repression, constitutive expression various control mechanisms, positive regulation, negative regulation, attenuation, operon hypothesis with special reference to mal/gal, ara and histidine operons, Regulatory mechanisms in bacteriophage lambda.

Regulation of gene expression at various levels (transcription, post transcriptional and translational)

DNA-protein interactions: Lambda family of repressor, trp repressor.

### Unit II: rDNA technology

Genomic and cDNA libraries, DNA manipulation enzymes, isolation of specific genes.

Gene cloning: REs, vectors-plasmids, cosmids phage vectors, M13 phage vectors, phagemids expression vectors with strong promoters, inducible, vectors produce fusion proteins and their isolation, Eucaryotic expression system, shuttle vectors, YAC, BAC insertion of DNA and its ligation to carrier DNA, introduction of DNA in cells, gene synthesis, gene libraries.

Application of recombinant DNA technology in medicine, agriculture industry and environmental sciences.

## **Unit III: Biochemical engineering**

Biochemical Engineering: Bioreactors and related equipment and instrumentation, types of bioreactor (Batch, semi batch, CSTF, recycle etc), reactor analysis, reactor design, reactor for recombination proteins.

#### **Unit IV: Fermentation technology**

Fermentation technology, microbial culture reaction, genetic modification, use of mutants, recombinant DNA technology and application in fermentation technology, microbial growth kinetics, sterilization, fermentation process kinetics, analysis of rate pattern and kinetic groups, fermentation process types, control of environmental variables, recovery of fermentation products, isolation and purification and use of immobilization techniques.

- 1. Molecular Biology of the Cell: Alberts 5<sup>th</sup> Edition 2007 NCBI Publication
- 2. Principles of Biochemistry: Lehninger WH Freeman
- 3. Biochemistry of Signal Transduction and Regulation Gerhard Krauss Wiley VCH 3<sup>rd</sup> Revised Edition
- 4. Molecular Cell Biology: Lodish 6<sup>th</sup> Edition, WH Freeman & Company
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- 7. Cell and Molecular Biology: Gerald Karp
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- 9. Molecular Biology of the Gene: Watson 6<sup>th</sup> Edition, Pearson Publication
- Gene Regulation: A Eukaryotic Perspective: David Latchman 5 illustrated , Taylor & Francis, 2005 Molecular Biotechnology: Principles and Applications of Recombinant DNA <u>Bernard R.</u> <u>Glick</u>(Author), Publisher: Amer Society for Microbiology; 4 edition (December 31, 2009)
- 11. Principles of gene manipulation and genomics <u>S. B. Primrose</u>, <u>Richard M. Twyman</u> Publisher: Wiley-Blackwell; 7 edition (February 17, 2006)
- 12. Principles of Fermentation Technology, <u>P. F. Stanbury</u> (Author), <u>S. Hall</u> (Author), <u>A. Whitaker</u> (Author) Publisher: Butterworth-Heinemann; 2 edition (February 19, 1999)

## BCH3T011: IMMUNOBIOLOGY

### **Unit I: Introductory Immunobiology**

### Complement system: Alternative and Classical pathway of complement activation

**Immune networks:** Homeostasis in the immune system-termination of normal immune responses, network hypothesis

In vivo immunity to viruses, bacteria, fungi, protozoa, worms etc

### Unit II: Immunological tolerance and Autoimmunity

Immunologic tolerance, T lymphocyte tolerance- central and peripheral, Apoptosis in Lymphocytes-pathways and biochemical mechanisms, effector mechanisms, Tolerance induced regulatory T cells, B lymphocyte tolerance-Central and Peripheral, Homeostasis in the immune, pathogenesis and therapeutic approaches to autoimmunity.

### Unit III: Tumor and Transplantation Immunology, Hypersensitivity

General features of tumor immunity, tumor antigens, Immune response to tumor and evasion, Immunotherapy, Types of hypersensitivity, Effector mechanisms of immunologic tissue injury and disease.

#### Unit IV: Immunodeficiency and Vaccinology

MHC and disease susceptibility, immune deficiency disorders, Active immunization (immune prophylaxis), passive immunization, adjuvants, modern approaches to vaccine development, role of vaccines in the prevention of disease.

- 1. Cellular and Molecular Immunology- 5<sup>th</sup> Edition, Abul K. Abbas, Andrew Litchman
- 2. Immunology-5<sup>th</sup> Edition, Richard A Goldsby, Thomas J. Kindt, Barbara A Osborne, Janis Kuby
- 3. Immunology- 6<sup>th</sup> Edition, Ivan Roitt, Jonathan Brostoff, David Male

# **BCH3T012: BIOCHEMICAL AND ENVIRONMENTAL TOXICOLOGY**

### Unit I: General principles of Toxicology

Definition, Different facets of toxicology and their interrelationships, Classification of toxic agents. Desired and undesired effects.

Various factors affecting toxicity: vehicles, formulation factors, biological half life, volume and concentration, dose, dosage forms, routes of administration / entry, genetic status etc.

Principles of selective toxicity: comparative morphology, comparative biochemistry, comparative cytology.

Toxicity assessment: acute, subchronic, chronic exposure, determination of ED50 and LD50 values, tests for mutagenecity, carcinogenicity, genotoxicity, Ames test.

### **Unit II: Disposition of Toxicants**

Factors affecting disposition of toxicants: absorption, distribution, biotransformation, elimination.

Absorption through gastro-intestinal tract, lungs, skin.

Distribution: storage in tissues, blood-brain barrier, passage across placenta, redistribution.

Biotransformation, Phase I and II reactions, metabolic interrelationship, antidotal therapy.

Excretion: urinary, fecal, exhalation, other routes.

Toxicokinetics: classic and physiologic.

#### Unit III: A) Non –organ directed toxicity

Chemical carcinogenesis: definition, mechanism.

Genetic toxicology: definition, health impacts and mechanism of induction of genetic alterations.

Developmental toxicology: definition, principles, mechanism and pathogenesis of developmental toxicity.

## **B)** Environmental Toxicology

Air pollution: definition, air pollutants, health effects and risk assessment of air pollution.

Introduction to Ecotoxicology

#### Unit IV: Target organ toxicity

Skin: skin as a barrier, dermatitis, acne, urticaria

Toxic responses of the blood: blood as a target organ, toxicology of erythron, leukon and platelets.

Toxic responses of the liver: physiology and pathophysiology, factors in liver injury, mechanism of liver injury.

Toxic responses of the respiratory system: lungs structure and functions, pulmonotoxic agents, pathogenesis of chemical induced damage, acute and chronic responses of lungs to injury.

- 1. Casarette and Doull's Toxicology by Klaassen CD
- 2. Biochemical Toxicology of Environmental Agents by Bruine D.
- 3. Detoxification mechanisms by Williams RT
- 4. Selective Toxicity by Albert A.
- 5. Developmental Toxicology by Hood RD.

# BCH3LAB5: BIOTECHNOLOGY AND IMMUNOLOGICAL TECHNIQUES

- 1) Fermentation
  - i) Isolation of microorganisms from soil demonstrating synthesis capability of desired product, Gram staining (Desired property to be decided by the instructor), and screening
  - ii) Optimization of the lab scale production of the desired product: Effect of temperature, pH, substrate concentration
  - iii) Growth curve: Estimation of cell number, substrate utilization and/or product formation.
- 2) Polymerase Chain Reaction
  - i) PCR amplification from genomic DNA
  - ii) Nested PCR
  - iii) Random Amplification of Polymorphic DNA (RAPD)
- 3) Restriction Fragment Length Polymorphism (RFLP)
- 4) Immunology
  - A. Quantitative Estimation of Antibody
  - B. Precipitation Techniques : Double Immunodiffusion, Single (Radial) Immunodiffusion
- 5) Electrophoretic Techniques : Immuno-eelectrophoresis, Rocket Immunoelectrophoresis, Immuno-diffusion

# BCH3LAB6: BIOCHEMICAL AND ENVIRONMENTAL TOXICOLOGY

- Qualitative detection of various toxicants in biological samples: Phenothiazine derivatives, Organochlorine compounds (Fujiwara test), Phenol, Methanol, Arsenic (As), Antimony (Sb), Selenium (Se), Mercury (Hg), Bismuth (Bi), Fluoride (F), Boron (Bo), Gutzeit test for Antimony (Sb) and Arsenic (As), Spot test for metal toxicants.
- 2. Quantitative determination of Salicylate, Paracetamol (acetaminophen), Sulphonamide in biological samples.
- 3. Enzyme assay in toxic conditions: GOT (AST), GPT (ALT), Acid phosphatase, Alkaline phosphatase, Acetyl cholinesterase etc.
- 4. Construction of dose-response curves.
- 5. Determination of LD50 value of a toxicant.
- 6. Induction of hepatotoxicity / diabetes / skin lesions / teratogenesis.
- 7. Organ / tissue morphology / histopathology
- 8. Assay of toxicant biotransformation enzyme-cytochrome P450.
- 9. Test for teratogenecity / carcinogenecity / Ames test.
- 10. Assay of biomarkers of environmental pollution / toxicity.

## BCH3INT3: INTERNAL ASSESSMENT: PRE-PROJECT PRESENTATION FOR APPROVAL