GOVT. DEGREE COLLEGE BARAMULLA (AUTONOMOUS)

SEMESTER 4th (NEP)

Major/Monor

SUBJECT: BIOTECHNOLOGY

Title: (BIOTECHNIQUES)

CREDIT: (4+2) THEORY: 04; PRACTICAL: 02

Code: BTGC1422M CONTACT HOURS: 64 (T) + 64 (L)

Objective: The major objective of this paper is to develop understanding of the key concepts of various techniques used across biological sciences, with a focus on principle and design of the instruments. This will enable the students to connect between theoretical concepts of these techniques and their immense biological applications in diverse fields.

Expected Learning Outcomes: Upon successful completion of the course, the student:

- 1. Will have identified the principle components of a microscope, simultaneously learning about the principles and practical applications in visualizing, identifying and measuring cell and its components.
- 2. Will be familiar with staining and preparation of samples for microscopy.
- 3. Will be acquainted with the working of spectrophotometer and will also gain the knowledge about its various applications.
- 4. Will have clear fundamentals of centrifugation, different types of rotors used, principle and working of differential and density gradient centrifugation, and the specific uses of ultracentrifuge.
- 5. Will have gained an in-depth knowledge of principles and applications of different types of chromatography.
- 6. Will have learnt basic concepts of various techniques like agarose gel electrophoresis, native polyacrylamide gel electrophoresis, SDS PAGE, isoelectric focusing, 2D gel electrophoresis

Unit - I Microscopy: 16 Hours

Principle, working and applications of light microscopy - bright-field, dark-field, phasecontrast,fluorescence & confocal microscopy, electron microscopy - TEM and SEM; Staining - principle and procedure of simple staining, negative staining & differential staining; Spectrophotometer- components of spectrophotometer, Beer-Lamberts law.

Unit - II Centrifugation and Chromatography: 16 Hours

Basic principle and applications of preparative and analytical centrifugation (differential centrifugation & density-gradient centrifugation) Types of centrifuges and rotors.

Chromatography-Introduction to the principle of chromatography.Paper chromatography, thin-layer chromatography, gel filtration, affinity and ion-exchange chromatography, HPLC.

Unit -III Electrophoresis: 16 Hours

Electrophoresis- general principle and types; Principle, procedure and applications of native polyacrylamide gel electrophoresis, sodium dodecyl sulphate-polyacrylamide gel electrophoresis, isoelectric focusing, two- dimensional gel electrophoresis and agarose gel electrophoresis.

Unit -IV Immunological techniques: 16 Hours

Principle, procedure and application of immunodiffusion, immuno-electrophoresis, enzyme linked immunosorbent assay (ELISA) and radioimmunoassay (RIA); **Radioisotope techniques:** Concept of radioisotopes, types and properties of radioactive decay, units of radioactivity, characteristics of radioisotopes commonly used in biology, measurement of radioactivity.

PRACTICALS (2 CREDITS)

- 1. Hands-on training on operation and use of microscope
- 2. Demonstration of protein purification techniques using paper chromatography and HPLC
- 3. Protein extraction and purification from mammalian/bacterial/plant cells
- 4. Separation of proteins through SDS-PAGE
- 5. Estimation of DNA by diphenylamine
- 6. Preparation of Agarose gel, and Electrophoresis of DNA
- 7. Demonstration of ELISA and Immunodiffusion

SUGGESTED READINGS

- Karp, G. 2010. Cell and Molecular Biology: Concepts and Experiments. 6th Edition. John Wiley& Sons. Inc.
- 2. *Principles and Techniques of Biochemistry and Molecular Biology*: Wilson, K. and Walker, J. Cambridge University Press.
- 3. *Physical Biochemistry Applications to Biochemistry and Molecular Biology:* Freifelder, D. - W. H.Freeman and Company.
- 4. A manual of "Introductory Practical Biochemistry", (2000), S. K. Sawhney, Randhir Singh Narosa, 2000.
- 5. Practical Enzymology, 2nd edition (2011), Hans BissWanger, Wiley Blackwell, USA
- 6. Biophysical chemistry Principle and techniques; Upadhyay, Upadhyah, Nath: Himalaya Publishinh House

GOVT. DEGREE COLLEGE BARAMULLA (AUTONOMOUS)

SEMESTER 4th (NEP)

MAJOR

SUBJECT: BIOTECHNOLOGY

Title: (CELL SIGNALLING AND CANCER BIOOLOGY)Code: BTGC3422MCREDIT: (4+2) THEORY: 04; PRACTICAL: 02CONTACT HOURS: 64 (T) + 64 (L)

Course Objective: This course aims to empower the learners by providing understanding of interaction of cells with other cells. It will provide deep understanding of cellular aspect of mechanism of signal transduction, cell cycle and cancer.

Expected Learning Outcomes: Upon successful completion of the course, the students:

- 1. Will be familiarized with the interaction of cell with the extracellular matrix.
- 2. Will have gained in-depth knowledge about signal transduction pathways with understanding of different type of receptors and signaling molecules.
- 3. Will be familiarized with the events of cell cycle, cell division and cell death and deep understanding of events of mitosis, apoptosis.
- 4. Will have acquired knowledge about biology of cancer its causes and different therapeutic approaches in controlling cancer.

Unit – I: Extracellular Matrix (16 Hours)

Extracellular Matrix: composition, molecules that mediate cell adhesion - role of integrins, focal adhesion and hemidesmosomes. Cell to cell interaction and roles of different adhesion molecules: selectins, immunoglobulin superfamily, cadherins. Adherens junctions and Desmosomes, Tight junctions, Gap junctions and Plasmodesmata

Unit – II: Cell Cycle (16 Hours)

Cell cycle: stages of cell cycle, role of cdks, control of cell cycle: check points and cdk inhibitors, overview of M phase: mitosis and cytokinesis, overview of meiosis. Programmed cell death (apoptosis), apoptosis vs necrosis, brief idea of autophagy

Unit – III: Signalling (16 Hours)

Signaling - autocrine, paracrine and endocrine signaling; extracellular messengers and their receptors. Signal transduction through cell surface receptors: GPCR signaling pathway, Receptor tyrosine kinase pathway, Ras-MAP Kinase Pathway, Signal transduction through intracellular receptors, Secondary messengers: role of cAMP, cGMP, NO, calcium, IP3, DAG as intercellular second messengers

Unit – IV: Cancer (16 Hours)

Cancer: basic properties of cancer cell, causes of cancer, genetic rearrangements in progenitor cells, oncogenes, tumor suppressor genes, cancer and the cell cycle, virus-

induced cancer, metastasis, interaction of cancer cells with normal cells, therapeutic interventions of uncontrolled cell growth

PRACTICAL/TUTORIALS (2 CREDITS)

- 1. Presentations on different topics
- 2. Review paper writing
- 3. Visit to State Cancer Research Institute SKIMS Soura.

BOOKS RECOMMENDED

- 1. *Molecular Biology of the Cell*: Alberts, B., Bray, D., Lewis, J., Raff, M., Roberts, K. and Watson, J.D. Garland Publishing Inc. New York.
- 2. Cell and Molecular Biology Concepts and Experiments: Karp, G. John Wiley Inc. New York.
- 3. De Robertis, E.D.P. and De Robertis, E.M.F. (2006). Cell and Molecular Biology. VIII Edition. Lippincott Williams and Wilkins, Philadelphia.
- 4. Cooper, G.M. and Hausman, R.E. (2009). The Cell: A Molecular Approach. Edition. ASM Press & Sunderland, Washington, D.C.; Sinauer Associates, MA.
- 4. Becker, W.M., Kleinsmith, L.J., Hardin. J. and Bertoni, G. P. (2009). The World of the Cell. VII Edition. Pearson Benjamin Cummings Publishing, San Francisco.

GOVT. DEGREE COLLEGE BARAMULLA (AUTONOMOUS)

SEMESTER 4th (NEP)

SUBJECT: BIOTECHNOLOGY

Title: (RECOMBINANT DNA TECHNOLOGY)Code: BTGC2422MCREDIT: (4+2) THEORY: 04; PRACTICAL: 02CONTACT HOURS: 64 (T) + 64 (L)

Course Objectives: The objectives of the course are

- 1. To ensure that the student develops a clear comprehension of the concepts of recombinant DNA technology.
- 2. Acquaint students with the enzymes and cloning vectors used in RDT.
- 3. Creation of genomic and cDNA libraries and recombinant products.
- 4. Explore the applications of RDT in the synthesis of different recombinant products.
- 5. Comprehend the mechanism of transformation in competent cells and target DNA delivery.

Expected Learning Outcomes: Upon successful completion of the course, the students:

- 1. Will get familiarized with basic cloning tools such as enzymes used to manipulate DNA, and cloning vectors.
- 2. Will gather in-depth knowledge of DNA amplification and sequencing methods, and become conversant with construction and screening of genomic and cDNA libraries
- 3. Will have learnt various gene delivery methods and production of transgenic plant.
- 4. Will become aware of the applied aspects of all major techniques being used for the benefit of humankind.

Unit - 1: 16 Hours

Recombinant DNA technology tools - restriction endonucleases, ligases, phosphatases, T4 polynucleotide kinase, DNA polymerase I and Klenow fragment; Cloning vectors - general features of plasmids, bacteriophages (lambda & M-13), cosmids, phagemids; Basic cloning strategy, concept of linkers and adaptors, selection and screening of clones.

Unit - 2: 16 Hours

Gene libraries. Construction of Genomic and cDNA libraries, Screening of libraries by colony hybridization. PCR (polymerase chain reaction), Types of PCR- inverse PCR, reverse transcription PCR, hot start PCR, qPCR, applications of PCR. Site-directed mutagenesis. DNA fingerprinting by RFLP and RAPD. Hybridization and Blottin techniques. Gel retardation assays. DNA footprinting by DNase I.

Unit - 3: 16 Hours

DNA sequencing: chain termination DNA sequencing and automated sequencing. Human genome sequencing project, Rice genome sequence, Products of recombinant DNA technology: Human protein insulin, hGH and Factor VIII. Therapeutic products produced by genetic engineering- tPA, interferons, vaccines. Brief idea of CRISPR/Cas9.

Major Course

Unit - 4: 16 Hours

Methods of gene delivery in plants and animals: Microinjection, biolistic method (gene gun), electroporation, liposome mediated, chemical method and viral-mediated delivery, Agrobacterium-mediated gene delivery in plants: Role of Ti plasmid and its features, cointegrative and binary vectors, Transgenic plants with reference to Bt transgenic cotton, Golden rice, Antisense RNA technology.

PRACTICALS (2 CREDITS)

- 1. Isolation of genomic DNA
- 2. Isolation of plasmid DNA
- 3. Digestion of genomic/plasmid DNA using restriction enzymes and analysis by Agarose gel electrophoresis
- 4. Preparation of competent cells
- 5. Transformation of competent cells and screening of recombinants
- 6. Demonstration of southern blotting
- 7. Demonstration of PCR
- 8. A visit to a Research lab

SUGGESTED READINGS

- 1. Brown TA. (2006). Gene Cloning and DNA Analysis. 5th edition. Blackwell Publishing, Oxford, U.K.
- 2. Clark DP and Pazdernik NJ. (2009). Biotechnology-Applying the Genetic Revolution. Elsevier Academic Press, USA.
- 3. Glick, B.R., Pasternak, J.J. (2003). Molecular Biotechnology- Principles and Applications of recombinant DNA. ASM Press, Washington
- 4. Primrose SB and Twyman RM. (2006). Principles of Gene Manipulation and Genomics, 7th edition. Blackwell Publishing, Oxford, U.K.
- 5. Sambrook J, Fritsch EF and Maniatis T. (2001). Molecular Cloning-A Laboratory Manual. 3rd edition. Cold Spring Harbor Laboratory Press.
- 6. Genetic Engineering by P.K.Gupta
- 7. A manual of "Introductory Practical Biochemistry", (2000), S. K. Sawhney, Randhir Singh Narosa, 2000.