# DEPARTMENT OF BIOCHEMISTRY FACULTY OF ARTS, SCIENCE AND HUMANITIES KARPAGAM ACADEMY OF HIGHER EDUCATION

(Deemed to be University, Established Under Section 3 of UGC Act 1956) Eachanari PO, Coimbatore – 641 021, India.

# M.Phil / Ph.D– BIOCHEMISTRY (Effective from the academic year 2016- 2017 and onwards)

# PREAMBLE

- The degree of Master of Philosophy [M.Phil] /Doctor of Philosophy (Ph.D) is awarded to a candidate who has submitted a thesis on the basis of original and independent research in any biochemistry field of research.
- This makes a contribution to the advancement of knowledge, which can be useful to the society.

# DEPARTMENT OF BIOCHEMISTRY FACULTY OF ARTS, SCIENCE AND HUMANITIES RESEARCH PROGRAM – M.Phil / PhD in Biochemistry (2016–2017 and onwards)

Course code	Name of the course	Instruction hours / week	credits	Maximum Marks (100)
16RBC101	Research Methodology and Pedagogy	4	4	100
16RBC201	Advanced paper in Biochemistry	4	4	100
16RBC301A	Enzyme and Enzyme technology	4	4	100
16RBC301B	Cancer Biology and immunology			
16RBC301C	Medicinal Plants and Plant therapeutics			
16RBC301D	Clinical Biochemistry and Toxicology			
16RBC301E	Plant Molecular Biotechnology			
16RBC301F	Animal Tissue Culture			
16RBC301G	Fish nutrition and tissue culture	1		
Program Total		12	12	300

 16RBC101
 RESEARCH METHODOLOGY & PEDAGOGY

 4H-4C

Instruction hours/week: L: 4 T: 0 P: 0

Marks: 100 End Semester Exam: 3 Hours

#### **Course Objective:**

### Equip the students with:

- The availability of tools for literature search
- Usage of tools and documentation of available research
- Identification of lacunae in the literature
- Processing of data and evaluating statistical significance
- Nuances of scientific writing
- Dissemination of research data

#### **Course Outcome:**

#### After successful completion, the students will understand

- 1. Scopus, Cochrane, Pubmed databases
- 2. Collation of available research and exportation to appropriate format
- 3. Methods involved in the identification of lacunae in the literature
- 4. How to Process data and evaluate statistical significance
- 5. Nuances of scientific writing
- 6. Dissemination of research data

# UNIT- I

Fluorescent Antibody assay – Histochemical localization, ELISA techniques – Principles & Applications. Immuno radiometric assay – Principles & Applications Natural Products – Detection of bioactive molecules by gas chromatography, HPLC and HPTLC , Mass spectrometry, NMR. Emission Spectroscopy – Fluorescence, Phosphorescence and Chemiluminescence. X-Ray diffraction and Flow injection analysis.

#### UNIT- II

Flow Cytometry – Principles, Abnormal chromosome analysis, Karyotyping, Comet assay. DNA Fragmentation analysis – Microfabrication techniques and uses in biological applications. PCR methodology – Design of primers – RTPCR. PCR in genomic analysis and diagnostic applications. PFGE – Principles, techniques and applications.

#### UNIT -III

Biological database- DNA sequence database, protein sequence database SRS – Similarity searching, BLAST, FASTA Local and Global alignment, Multiple sequence alignment – Phylogeny. Structure database – Secondary structure prediction. Predicting 3D folds (Threading) Visualisation tool.

## UNIT-IV

Scientific writing – writing methodology, results & discussion, presentation. Correlation & regression. Sampling distribution – Students t test

Experimental design – CRD, RBD. Analysis of experimental results – ANOVA and its interpretation. Duncan's Multiple Range Test.

**Microsoft Windows**: Macintosh versions, **Microsoft Word** - Characteristics - Document statistics- Typical usage, **Microsoft Excel** - Basic operation – Charts - Using other Microsoft applications - Using external data, **Microsoft Power Point** – power point viewer – versions – uses, **Microsoft Access** – Uses – Features.

#### UNIT- V

#### Pedagogical methods in higher education

Objectives and role of higher education- Important characteristics of an effective lecture- Quality teaching and learning- lecture preparation- characteristics of instructional design- Methods of teaching and learning: Large group- Technique – lecture, seminar, symposium, team teaching, project, small group technique-simulation, role playing demonstration, Brain storing, Case discussion, and assignment, methods of evaluation- self evaluation, student evaluation, diagnostic testing and remedial teaching- question banking- electronic media in education:- 'e' learning researches- web based learning.

- Adrian Slater, Neigel Scott and Maark Fowler (2008), Plant Biotechnology Genetic manipulation of plants, 2<sup>nd</sup> edition, Oxford University Press, New York
- 2. Akay M. (Ed) Genomics and Proteomics Engineering in Medicine and Biology 2007 Wiley- Interscience John Wiley & sons, Inc. Publication, USA.
- 3. Deepak Bhariog (2006). Fundamentals of Information Technology, 3<sup>rd</sup> Edition Excel Books India.
- 4. Hall, Christopher; Tews, Carey (Retrieved 7 November 2010). "Mac Office matches Windows almost". InfoWorld. p. 117.
- 5. Hung T. Nguyen (2006), Fundamentals of mathematical statistics, Springer Berlin Heidelberg publishers.
- 6. Gupta, S.P. (2008), Statistical methods Sultan Chand & Sons. New Delhi

- Lodish H., Berk A., Kaiser CA., Kriger M .,Scott M.P.,Bretscher A., Ploegh H., Matsudaira P (2008) Molecular Cell Biology,6<sup>th</sup> edition, W.H.Freeman and Company, New York.
- Sensen C.W. [Ed.] (2007) Essentials of Genomics and Bioinformatics. Wiley-VCH, 419 pages. ISBN 978-3527612642

#### 2016-2017

16RBC201ADVANCED PAPER IN BIOCHEMISTRY

#### **4H-4C**

Instruction hours/week: L: 4 T: 0 P: 0

### Marks: 100 End Semester Exam: 3 Hours

### **Course Objective:**

### Equip the students with:

- Applications and limitations of antibody based assays
- Applications and limitations of fluorescence based assays
- Applications and limitations of microscopic assays
- Applications and limitations of nucleic acid analysis
- Analysis of protein using wet lab and computational methods
- Basic methodology of animal and plant tissue culture methods

### **Course Outcomes:**

### After successful completion, the students will understand

- 1. Appropriate antibody based assays for a specific research question
- 2. Appropriate f fluorescence based assays for a specific research question
- 3. Specific microscopic assays for a research question
- 4. Appropriate nucleic acid analysis for a specific research question
- 5. Usage of wet lab and computational methods for a specific research question
- 6. General methodology of animal and plant tissue culture methods

# UNIT I

#### Metabolism

Carbohydrate metabolism: TCA Cycle, Glycolysis, HMP, Gluconeogenesis, Lipid Metabolism: Fatty acid synthesis and Beta oxidation and Chain elongation Nucleic acid metabolism: Purine and Pyrimidine metabolism

Protein metabolism: Urea cycle, Aliphatic and aromatic amino acid metabolism

# UNIT II

# Cell Biology and Molecular Biology

Replication,transcription and translation in pro and eukaryotes. RNA editing, miRNA and RNAi and its applications. Antisense RNA Technology,signal sequence hypothesis.

General principle of cell communication- G-protein coupled receptor-structure and functions ,cyclic AMP and other second messengers-Phosphatidyl inositol, Diacyl glycerol, Inositol 1,4,5 triphosphate,Ca<sup>2+</sup>;receptor tyrosine kinases-structure and functions, importance of Ras, MAP kinase cascade.

# UNIT -III

#### **Genomics and Proteomics**

Human Genome project-History, techniques and applications: Anatomy of prokaryotic and human genome: genetic mapping and genetic markers-RFLP, Mini and micro satellite, STS and EST, SSCP, RAPD, AFLP, SNPs, Analysing gene expression-DNA micro array.

Proteome analysis- 2D gel electrophoresis: protein-protein interactions- yeast twohybrid system and protein micro arrays.

### UNIT -IV

### **Plant Biotechnology**

Tissue culture media, composition and preparation, primary culture, callus and suspension cultures, somoclonal variation, micro propagation, organogenesis, Somatic embryogenesis, artificial seeds, Transfer and establishment of whole plants in soil, Haploidy: Protoplast fusion and somatic hybridization.

### UNIT –V

#### **Animal Biotechnology**

Media: Natural media, balanced salt solution and simple media, serum and protein free chemically defined media. Primary cell culture,(chick, mouse and human biopsy)and methods of desegregations of tissues; continuous or established cell culture, tissue culture, organ culture; three dimensional culture, feeder layer, cell separation; cell synchronization; cryopreservation and revival.

- 1. Alberts, B. et al. (2008). Molecular Biology of the Cell. 5th Ed. Garland Publishing House, USA.
- 2. Becker. (2009). The World of the Cell. 7th Ed. Benjamin-Cummings, USA.
- 3. Clark, D. P. (2005). Molecular Biology. Elsevier. USA.
- 4. Gupta P.K. (2010), Biotechnology & Genomics, 5th Reprint, Rastogi Publications Meerut.India
- 5. Freshney, R. I., & Freshney, M. G. (2010). In Freshney, R. I. (ed.), Animal cell culture: a practical approach, 2nd ed. IRL Press at Oxford University Press.
- 6. Karp G. (2012), Cell and Molecular Biology: Concept and Experiments. John Willy, New York, USA.
- 7. Lodish et.al. (2013) Molecular Cell Biology, 7<sup>th</sup> edition, W.H.Freeman and Company, New York.USA.
- 8. Watson, J. D., Baker, T. A. & Bell, S. P. (2007). Molecular Biology of the Gene. 6th Ed. Benjamin Cummings.USA.

### 16RBC301A ENZYMES AND ENZYME TECHNOLOGY 4H-4C

# Instruction hours/week: L: 4 T: 0 P: 0

#### **Course Objective:**

#### Equip the students with:

- Techniques involved in the analysis of protein structure and function
- The applications and limitations of methods involved in isolation of enzymes
- The applications and limitations of methods involved in purification of enzymes
- Physical, Chemical and Biological methods of enzyme immobilization
- Clinical significance of enzyme analysis
- Biotechnological applications of enzymes

#### **Course Outcomes:**

### After successful completion, the students will understand

- 1. X-Ray crystallographic, Spectrophotometric analysis of enzymes
- 2. How to estimate the fold purity during isolation of an enzyme
- 3. Enzyme purification techniques
- 4. Physical, Chemical and Biological methods of enzyme immobilization
- 5. The clinical significance of enzyme analysis in analyzing the vital organ functions

Applications of enzymes in the area of medical and industrial biotechnology

# UNIT- I

### Protein and enzymes

Protein structure, functions, compositions and conformation of proteins. Enzyme catalysis-Acid base catalysis, covalent catalysis, an example, serine proteases. Enzyme kinetics – Michaelis menton equation, Line weaver Burk plot, Hills equation, Hans plot.

#### UNIT -II

#### Isolation and purification of enzymes

Sources of enzymes for industry, extraction of enzymes for scientific and industrial purposes. Downstream processing of enzymes, uses of soluble enzymes. Study of enzymes in aqueous biphasic systems. Factors affecting the enzyme activity - Substrate concentration, Enzyme concentration, pH, temperature etc.,

#### UNIT-III

#### Enzyme immobilization and their applications.

Techniques employed for immobilizing enzymes, kinetics of immobilized enzymes. Advantages and disadvantages in the utilization of soluble enzymes, Immobilized enzymes and immobilized cells. Different types of reactors of immobilized enzymes and their applications.

#### Marks: 100 End Semester Exam: 3 Hours

# UNIT -IV

### Clinical analysis of enzymes

Application of ELISA and EMIT in clinical analysis. Different types of Biosensorspotentiometric, amperometric, piezo - electric and immuno biosensors. Electro analytical applications of enzymes, Methods of coenzyme regeneration. Biochips and Biocomputers.

# UNIT -V

## **Enzymes in Biotechnology**

Enzyme catalysis in organic solvents, Restriction endonucleases, DNA ligases, DNA polymerase and their uses in Biotechnology. Site directed mutagenesis, artificial enzymes, ribozymes and Abzymes and their uses.

- 1. Bommarius A.S., B.R. Riebel. 2004. Biocatalysis Fundamentals and Applications, Wiley-VCH, Weinheim, Germany.
- 2. Buchholz K., V. Kasche, U.T. Bornscheuer. 2005. Biocatalysts and Enzyme Technology, Wiley-VCH, Weinheim, Germany.
- 3. Cook P. F., W.W. Cleland. 2007. Enzyme Kinetics and Mechanism, Garland Science Publishing, London, England and New York, USA.
- 4. Irwin Segel. 2004. Biochemical Calculations, John Wiley and Sons, California, USA.
- 5. Marangoni A.G. 2003, Enzyme Kinetics-A Modern Approach,
- 6. Nicholas C. 2004 Fundamentals of Enzymology. Third Edition. Price and Lewis Stevens. USA.
- 7. Palmer T. 2001 Enzymes Biochemistry, Biotechnology and Clinical Chemistry, 5th Edition, Howood Publishing Chishester, England.
- 8. Satyanarayana, U., 2006. Biotechnology, Books and Allied (P) Ltd. India.
- http://biotech buddy. google pages.com/enzyme technology.html.
- http://www.Isbu.ac.uk? biology/enztech.
- http://www.Science direct.com/science
- http://www.woodhead publishing.com/en/book.asp.

### 16RBC302B CANCER BIOLOGY AND IMMUNOLOGY 4H-4C

#### Instruction hours/week: L: 4 T: 0 P: 0

Marks: 100 End Semester Exam: 3 Hours

#### **Course Objectives:**

#### Equip the students with:

- Etiology of cancer
- Steps involved in the progression of cancer
- Changes in the cell division during oncogenesis
- Programmed cells death
- Immune surveillance strategy
- Techniques involved in the assessment of cancer development and progression

#### **Course Outcomes:**

#### After successful completion, the students will understand:

- 1. The reasons for the development of cancer
- 2. Factors involved in the advancement of cancer
- 3. Targeting the cell division for the treatment of cancer
- 4. Targeting apoptosis for the treatment of cancer
- 5. Methods of enhancing immune surveillance for cancer treatment
- 6. Usage of appropriate techniques for assessment of cancer stages

#### UNIT I

Biology of cancer-Phenotype of a cancer cell causes of cancer-DNA tumor viruses, RNA tumor viruses, cell cycle and its control-role of protein kinases, checkpoints, kinase inhibitor and cellular response.

#### UNIT II

Programmed cell death (Apoptosis)-Intracellular proteolytic cascade, cascade of caspase proteins, adapter proteins, Bcl-2, IAP family proteins, extra cellular control of cell division, tumor necrosis factor and related death signals.

#### UNIT III

Genetic basis of cancer-oncogenes, tumor suppressor genes, aberrations in signaling pathways. oncogenic mutations in growth promoting proteins, Mutations causing loss of growth –inhibiting and cell cycle control, Role of carcinogens and DNA repair in cancer.

#### UNIT IV

Immunity- Active, passive, humoral and cell mediated immunity. Therapeutic uses of cytokines and cytokine receptors. Test for lymophocyte function. B cell and T cell

immuno deficiency disorder. Clinical laboratory methods for the detection of antigens and antibodies test for histocompatability antigens, neoplasm of the immune system.

# UNIT V

Techniques-FISH techniques, Real time PCR, Western blotting, ELISA assay, immunocytochemistry, immunohistochemistry, flow cytometry, fluroscent microscopy and confocal microscopy.

- 1. Alberts, B. et al. (2008). Molecular Biology of the Cell. 5th Ed. Garland Publishing House. USA.
- 2. Benjamin Lewin (2007) Genes VIII, Prentice Hall. USA.
- Brown T.A. (2010), Gene Cloning & DNA Analysis, 6nd Edition, Wiley-Blackwell, New York.
- 4. Karp G. (2012), Cell and Molecular Biology: Concept and Experiments. John Willy, New York.
- 5. Klug, W.S., Cummings, M.R, Spencer C.A and Palladino, M.A. (2012), Concept of Genetics, 10th Edition, Pearson Education, Singapore.
- Lodish H., Berk A., Kaiser CA., Kriger M .,Scott M.P.,Bretscher A., Ploegh H., Matsudaira P.2008. Molecular Cell Biology, 6<sup>th</sup> edition, W.H.Freeman and Company, New York.
- 7. Janeway et al., 2012.Immunobiology, 8th Edition, Current Biology publications, USA.
- Watson J.D. 2009, A Passion for DNA: Genes, Genomes & Society, Cold Spring Harbor Laboratory press (CSHL)

2016-2017

# 16RBC303C MEDICINAL PLANTS AND PLANT THERAPEUTICS 4H-4C

### Instruction hours/week: L: 4 T: 0 P: 0

## Marks: 100 End Semester Exam: 3 Hours

### **Course Objectives**

### Equip the students with:

- Methods to identify plants/herbs with a specific biological activity
- Evaluating the presence/absence of flavonoids, terpenoids, glycosides and steroids
- Analysis of the crude extract in vitro to evaluate its free radical scavenging activity
- Fractionation methods to isolate a specific compound and its evaluation
- Enhancing the production of secondary metabolite using plant tissue culture
- Assessment of the efficacy of a specific plant metabolite using animal cell culture

### **Course Outcomes:**

# After successful completion, the students will understand:

- 1. The availability of traditional knowledge search resources
- 2. Identify flavonoids, terpenoids, glycosides and steroids
- 3. Analyze the crude extract in vitro for its free radical scavenging activity
- 4. Column chromatography, TLC
- 5. How to enhance the production of secondary metabolite using plant tissue culture
- 6. Animal cell culture

# UNIT I

Medicinal plants-bioactive principles in medicinal plants methods of extraction, isolation, separation and screening, pharmacologically active plants-CNS, CVS, Hypoglycemic, Hepatoprotective, anti allergic, anticancer, immunoactive plants, plants protecting against oxidative stress, chemotherapeutic products.

# UNIT II

Free radicals –types, sources, importance, production, free radicals induced damages, lipid peroxidation, measurement of free radicals, disease caused by radicals, reactive oxygen species, antioxidant defence system, enzymic and nom-enzymic antioxidants, role of antioxidants in prevention of diseases, phytochemicals as antioxidants.

# UNIT III

Alkaloids, flavanoids, terpenoids, phenols-Occurrence ,distribution & functions ,Production of secondary metabolite in plants, stages of secondary

metabolite production, uses of tissue culture techniques, elicitation, biotransformation- production of pharmaceutical compounds.

### UNIT IV

Principles-callus, meristem and organ culture, culture methods, culture media & preparations ,plant regeneration, protoplast technology, micropropogation in plants, somatic embryogenesis, somoclonal selection.

## UNIT V

Animal cell culture: Culture media, Serum and protein free defined media and their application. Functions of different constituents of culture medium. Role of carbon dioxide, growth factors, glutamine in cell culture. Cell lines, primary culture and culture maintenance.

Experimental animals and Animal handling, Sacrification, collection of sample. Ethical issues for animal handling.

- Dubey R.C. 2009.Text book of Biotechnology, S. Chand & Company Ltd. New Delhi
- Freshney, R. I., & Freshney, M. G. (2010). In Freshney, R. I. (ed.), Animal cell culture: a practical approach, 2nd ed. IRL Press at Oxford University Press.USA.
- 3. Jain V. K2010.. Fundamentals of plant physiology, C. Chand and Company Ltd, New Delhi
- 4. Purohit.S.S. (2005) Agricultural Biotechnology, Dr.Updesh Purohit Publishers, Jodhpur.India
- 5. Singh.M.P and Panda.H (2005).Medicinal Herbs with their formulations, Daya Publishing House, NewDelhi

2016-2017

## 16RBC304D CLINICAL BIOCHEMISTRY AND TOXICOLOGY 4H-4C

#### Instruction hours/week: L: 4 T: 0 P: 0

#### Marks:100 End Semester Exam: 3 Hours

#### **Course Objectives:**

# Equip the students with:

- The collection of blood, serum and plasma
- Analyzing the inflammatory markers
- Clinical significance of enzyme assays
- Role of oxidative stress in physiological and pathological state
- Enzymatic and non enzymatic antioxidants
- Toxicological studies

#### **Course Outcome:**

### After successful completion, the students will understand:

1. The different anti-coagulants used for the isolation of plasma and its significance

- 2. Assessment of inflammation
- 3. Role of enzymes to predict vital organ functioning
- 4. Regulation of oxidative stress
- 5. Significance of endogenous antioxidant system
- 6. Principles and applications of histo-chemical analysis

# UNIT-I

**Clinical Enzymology:** Clinical significance of Phosphatases, transmainases, 5'nulceotidase, Gamma –glutamyl transferase, Lactate Dehydrogenase, Creatine Phospho kinase

Diagnostic enzymes in hepatobiliary disease, Atherosclerosis, Myocardial infarction, renal dysfunction. Cancer markers for oral, prostate, colorectal breast and GI tract cancer, oncofetal cancer markers.

#### UNIT-II

Formation of free radicals, autoxidation initiated by oxygen radicals, Influence of free radicals in metal toxicity. Free radicals and cancer .Oxidative process in tissue injury. Detection of free radicals and radical ions. Role of free radicals in diseases.

#### UNIT-III

**Enzymic antioxidants-** Chemistry, mechanism, antioxidant effect of SOD, catalase, Glulathione Peroxidase.

**Non Enzymic antioxidants-** source, chemistry, toxicity, biochemical functions, bioavailability, bioassays, Antioxidant effects of Vit A, Vit C, Vit E, glutathione and selenium.

**Trace elements** -Introduction, sources, biochemical functions of zinc, copper and magnesium & iron.

# **UNIT-IV**

**Medicinal plants-**bioactive principles in medicinal plants methods of extraction, isolation, separation and screening ,Pharmacologically active plants-CNS,CVS, Hypoglycemic, Hepatoprotective ,anti allergic ,anticancer, immunoactive plants, plants protecting against oxidative stress, chemotherapeutic products.

# UNIT-V

Effects of physiochemical and biological factors on heavy metal toxicity, toxic mechanism- Carcinogenesis, teratogenesis & immunotoxicity. Bioassays for heavy metal toxicity, pathological Histopathological examinations for heavy metal toxicity.

- 1. Chatterjee M.N. and Rana Sinde, (2006) Text Book of Medical Biochemistry, 6<sup>th</sup> Edition, Jaypee Brothers, Medical Publishers, New Delhi
- 2. Harper's Illustrated Biochemistry 200928th Edition McGraw Hill, Mumbai
- 3. Nelson and Cox 2005Principles of Biochemistry by, 4th Edition,. Mumbai
- 4. Devlin 2006.Biochemistry with Clinical Correlation, 6 th Edition, John Wiley & Sons, USA.
- 5. Ramnik Sood 2009. Medical Laboratory Technology,; Jaypee Brothers Medical Publishers, New Delhi
- 6. Tietz Fundamentals of Clinical Chemistry2008. 6th Edition, Elsevier, USA.
- 7. Voet D.and Voet J.2008.Biochemistry, 3rd Edition, J. Wiley & Sons, USA.

M.Phil/Ph.D Biochemistry

#### 2016-2017

16RBC305EPLANT MOLECULAR BIOTECHNOLOGY4H-4C

#### Instruction hours/week: L: 4 T: 0 P: 0

Marks: 100 End Semester Exam: 3 Hours

#### **Course Objectives:**

Equip the students with

- The organization of plant genome
- Molecular markers of plant tissues
- Growth regulators of plant
- Cloning strategies
- Transformation methods
- Manipulation of plant nucleic acids

### **Course Outcomes**:

After successful completion, the students will understand:

- 1. Organization of plant genome
- 2. Molecular markers of plant tissues
- 3. Growth regulators of plant
- 4. Cloning strategies
- 5. Transformation methods
- 6. Manipulation of plant nucleic acids

#### UNIT-I

**Plant genome**: Plant genome organization, structural features of a representative plant gene. Organization of chloroplast genome and mitochondrial genome. Molecular markers (AFLP,ISSR and RAPD) . plant tissue culture media, plant hormones and growth regulators in tissue culture, preparation of suitable explants. Micropropagation of plants - somatic embryogenesis, protoplast culture, somatic hybridization and synthetic seeds.

#### UNIT-II

**Cloning strategies** - Tools for cutting and joining of DNA; gene transfer techniques;Methods of selection and screening of recombinant DNA. Construction of genomic libraries and cDNA libraries - probe construction and labelling (radio and non-radio).Molecular mechanism of anti-sense technology - inhibition of splicing , disruption of RNA structure & capping - application of anti-sensing technology.

#### UNIT-III

**Gene regulation:** Inducible enzymes, regulatory mutations, repressor, operon, promotor, catabolic repression, repressible enzyme systems, control by attenuation, positive control, gene regulation in eukaryotes, transcriptional regulation, post transcriptional regulation, hormones & gene expression; viruses & gene expression, genetic control of pattern formation in plant development.

**Unit IV – Plant transformation technology**: Symbiotic nitrogen fixation in legumes by rhizobia - biochemistry and molecular biology. Binary vectors, Use of

35s & other promoters genetic markers methods of nuclear transformation viral vectors & their applications, Use of reporter gene, Particle bombardment ,Electroporation, Microinjection, Chloroplast transformation, Transformation of monocots, Transgene stability & gene silencing in Plant transformation.

### UNIT-V

**Plant manipulation and its applications:** Transgenic plants - for- biotic (weeds, insects, viruses, fungi and bacteria) and abiotic (drought, salt, temperature, poor soil quality and oxidative) stress tolerance.Production of secondary metabolites production. Molecular farming (improvement in protein, lipids, carbohydrates. Plant antibodies, vaccines, therapeutic proteins and active principles. Biofortication of important crops (rice and banana).

- 1. Altman A, Hasegawa P M . 2012 "Plant Biotechnology and agriculture. Prospect for the 21<sup>st</sup> century" Academic press,USA
- 2. Brown T. A. 2010. Gene Cloning and DNA Analysis: an introduction, 6<sup>th</sup> edition, Wiley-Blackwell Publisher, UK.
- 3. Chawla H.C. 2009 Introduction to Plant Biotechnology 3r<sup>d</sup> Edition, Oxford & IBH publication Pvt .Ltd, New Delhi.
- 4. Davies K. 2004. Plant Pigments and their Manipulation Annual plant reviews, vol 14 Blackwell Publication, UK
- 5. Glick and Paster mark, 2002. Molecular Biotechnology Principles and Applications in Recombinant DNA, Panima Publishing Co-operation, Banglore
- 6. Primrose S.B and R.M.Twyman. 2003. Principles of Genome Analysis.Blackwell Publishing, Oxford.
- 7. Slater A, Scott NW, Fowler MR. 2008 Plant Biotechnology: the genetic manipulation of plants, Oxford Press, UK
- 8. Winnacker E.. 2003. From Gene to Clones ; Introduction to gene technology, 4<sup>th</sup> edition, Panima Publisher, India

16RBC306F ANIMAL TISSUE CULTURE

### Instruction hours/week: L: 4 T: 0 P: 0

Marks: 100 End Semester Exam: 3 Hours

### **Course Objectives:**

### Equip the students with:

- Applications and limitations of cell based studies
- Aseptic methods to perform animal cell culture
- Types of cell culture techniques
- Assessment of cell proliferation
- Assessment of cell differentiation
- Scale up technologies

### **Course Outcomes:**

### After successful completion, the students will understand:

- 1. The need and circumstances for the cell based studies
- 2. Sterile working culture
- 3. Suspension and adherent cell culture
- 4. Thymidine incorporation, WST-1 Assays
- 5. Galactosidase staining, von-kossa, and alizarin red staining
- 6. Organ culture

# UNIT I

Introduction, importance, history of cell culture development, different tissue culture techniques including primary and secondary culture, continuous cell lines, suspension culture, organ culture, advantages and limitations medical/pharmaceutical products of animal cell culture-genetic engineering of animal cells and their applications. Risks in a tissue culture laboratory and safety - biohazards.

# UNIT II

Different types of cell culture media, growth supplements, serum free media, balanced salt solution, other cell culture reagents, culture of different tissues and its application. Facilities for animal cell culture-infrastructure, equipment, culture vessels. Biology and characterization of cultured cells-cell adhesion, proliferation, differentiation, morphology of cells and identification.

# UNIT III

Primary cell culture techniques - mechanical disaggregation, enzymatic disaggregation, separation of viable and non-viable cells. Mass culture of cells - manipulation of cell line selection - types of cell lines - maintenance of cell lines - immobilization of cells and its application - synchronization of cell cultures and cell division - production of secondary metabolites - biotransformation - Induction of cell

#### 4H-4C

line mutants and mutations - cryopreservation – germplasm conservation and establishment of gene banks.

# UNIT IV

Animal cell culture scale up: Scale up in suspension - stirrer culture, continuous flow culture, air-lift fermentor culture; Scale up in monolayer - Roller bottle culture, multi surface culture, multi array disks, spirals and tubes - monitoring of cell growth. Organ culture - whole embryo culture - specialized culture techniques - measurement of cell death.

# UNIT V

Tissue engineering: Design and engineering of tissues - tissue modeling. Embryonic stem cell engineering - ES cell culture to produce differential cells - Human embryonic stem cell research. Transgenic animals-transgenic animals in xenotransplantation

- 1. Butler. M. 2004. Animal Cell Culture and Technology, BIOS Scientific Publishers, Taylor and Francis Group. U. K.
- 2. Freshney, R. I., & Freshney, M. G. 2010. In Freshney, R. I. (ed.), Animal cell culture: a practical approach, 2nd ed. IRL Press at Oxford University Press.
- 3. Gupta P.K. (2010), Biotechnology & Genomics, 5th Reprint, Rastogi Publications Meerut.
- 4. Ranga M.M., Animal Biotechnology, (2007) Agrobios, India.
- 5. Satyanarayana, U., 2006 Biotechnology, Books and Allied (P) Ltd. India.